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MEMOIRS.

On the Embryo-sac *and* Development *of* Gymnadenia
conopsea. By H. Marshall Ward, Scholar of Christ's
College, Cambridge. With Plates I, II, and III.

The hitherto received account of the origin, mode of for-
mation, and homologies of the embryo-sac in Angiosperms
was based on the researches of Hofmeister,[1] for the most
part, and may be summed up to the following effect:—A
single cell situated in the apical part of the nucleus, and
usually the foremost of an axial row, enlarges, its nucleus
disappears, and a variable small number of free nuclei
appear in the protoplasm by free cell formation. Of these,
two are close to the apex of the enlarged cell, and are
known as "germinal" or "embryonic" vesicles, while a
very inconstant number of "antipodal" cells often, but not
always, forms in the lower end of the protoplasm. The
enlarged cell is the embryo-sac, and receives the pollen tube
at its apex, where one, or at times two, of the germinal vesi-
cles become fertilised by the contents of the tube. Schacht[2]
showed that one of the germinal vesicles acted as a
conductor between the tube and the fertile vesicle, or that
two acted as such towards a third germinal vesicle, and
were often marked by peculiar longitudinal striæ.

This "filiform apparatus" and fertile germinal vesicle
were regarded as the homologues of the corpuscula[3] of the
Gymnosperms by Strasburger and Pringsheim, and therefore
were regarded as representing rudimentary archegonia, in
accordance with Hofmeister's views as to the "rosette" or
neck-cells in Conifers.[4] The antipodal cells came to be regarded
as the last and fleeting representatives of the endosperm,
which arises in the embryo-sac of Coniferæ, &c., and which

[1] "Neue Beiträge," &c., in 'Abhandlung d. Konigl. sächs Gesellsch. d.
Wiss.,' 1859 and 1861.
[2] 'Jahrb. f. Wiss. Bot.,' 1857.
[3] R. Brown (1834), 'Misc. Bot. Works.'
[4] 'The Higher Cryptogams,' Ray Soc., 1862.

had been recognised by Hofmeister as the homologue of a true prothallium.[1] It was also generally admitted that the endosperm of Angiosperms had nothing to do with that of Conifers, but rather corresponds to what is found appearing late in the developing macrospore of Selaginella, and independent of the prothallium.[2]

Quite lately the necessity for a reconsideration of these points has been forced upon us by the researches of Strasburger,[3] of Warming,[4] and the papers of Vesque,[5] which clearly showed that in a great number of types the embryo-sac does not arise so simply as Hofmeister thought, and further, that the processes going on between its formation and the origin of an embryo are still more complex.

Before touching upon this or examining the different views of the authors named, I propose to state what is found to occur in *Gymnadenia conopsea*, one of the commoner Orchids of Europe. This has been partly done by Strasburger,[6] who has, however, more especially described the processes in *Orchis pallens* and *Monotropa hypopitys*. The later stages have not, to my knowledge, been before described.

Except where stated otherwise, the method employed has been to cut sections of the ovaries which have lain in absolute alcohol at least twenty-four hours, and in a slowly evaporating mixture of alcohol and glycerin at least twenty-four hours longer. In a few cases staining with fuchsine and acetic acid has been resorted to successfully. As is well known, the ovules arise from three parietal placentæ, as outgrowths of a few cells projecting into the cavity of the inferior ovary. Hofmeister traced the Orchid ovule to one cell of the surface, whence arose the idea that it is morphologically a trichome.[7]

At a period shortly before the flower bud is complete, transverse sections of the ovary reveal the young ovules as straight or very slightly curved structures, arising in considerable numbers from each placenta and directed at right angles to its surface. Each consists of an axial row of large polygonal cells, surrounded by one layer of somewhat smaller cells. In transverse section we see one large cell with five or six

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[1] Loc. cit.
[2] Cf. also Sachs' 'Text Book,' Eng. Trans.
[3] 'Ueber Befruchtung und Zelltheilung,' 1877.
[4] 'Ann. d. Sc. Nat. Bot.,' 1878.
[5] Ibid.
[6] Op. cit., and 'Die Gymnosp. und Angiosp,' 1879.
[7] 'Cf. also Sachs' 'Lehrbuch.'

around it. The cells are thin-walled, and full of slightly granular protoplasm, without any vacuoles, and each has a large spherical nucleus with a bright nucleolus in the centre.

At an early stage the terminal cell of the central series increases considerably in size, its protoplasm becomes very granular, and its nucleus remarkably bright. About the same time certain cells in a zone above the middle of the whole ovule divide by walls parallel to the periphery of the ovule and form the rudiment of the inner integument. At this period the ovule is represented in fig. 1, where, however, the protoplasm has been mostly removed by ammonic hydrate.

It will be useful to name the central series of cells the "axial row," and the large terminal cell of this row has been called the "embryo-sac mother-cell," since the embyro-sac apparently arises as a daughter-cell by division of this, as pointed out by Strasburger.[1] Reasons will be given later for considering the embryo-sac as possibly less simple than here stated.

As the ovule lengthens its curvature increases, the integument grows upwards, and the embryo-sac mother-cell becomes longer (fig. 2); this latter cell now divides by a transverse wall appearing at about one third its length from the top, and the smaller cell thus cut off appears like a cap sitting on the larger one below. The lower and larger cell is meanwhile growing, and tends to compress this cap-cell against the epidermal cells above. When it has reached a size about equal to that with which it started, the larger cell repeats the above division exactly as before, and so a second cap-cell is thrown off, soon to be forced up against the other by growth of its sister-cell below.

The specimen represented in fig. 3 shows this second division in progress, the characteristic "barrel figure" having been fixed by absolute alcohol. A similar stage is figured by Strasburger ('Angiosp. u. Gymnosp.,' Taf. vi, fig. 90), where, however, the division appears to follow more rapidly upon the first one. The division walls thus established are remarkably thick and bright, as if mucilaginous and swollen, and the appearance they present is not easily indicated in a drawing, especially at stages a little later, when the whole becomes more homogeneous as the lower cell compresses the upper ones.

Meanwhile the ovule has become decidedly anatropous, and the inner integument is already slowly closing in above; the outer integument has also appeared, by similar divisions

[1] Loc. cit.

of the outer cells of a zone, at the base of the inner integu-
ment (cf. figs. 2, 3, 4). The lower of the three cells pro-
duced by division of the "embryo-sac mother-cell" becomes
longer, and in its growth compresses, not only the two cap-
cells, but also the epidermal layer on all sides. It appears
to absorb the contents of these cells, and the exhausted
remains easily suffer compression to mere refractive homo-
geneous masses, as their greedy neighbour grows. Since
this cell apparently becomes the embryo-sac we shall here-
after speak of it as such; it has now again become about
as long as at first, and proportionately wider, and its granu-
lar protoplasm contains a large nucleus.

During the progress of the changes about to be described
the two integuments must be pictured as growing up, and
the inner one closely investing the nucleus and forming a
micropyle above; the outer integument never closes in so
much, but leaves a large loose opening above in later stages.
Not many cells are formed, but as the embryo-sac becomes
completed the integument cells elongate very much, large
sap cavities form, and the nuclei are driven to their walls.
This is especially the case with the outer integument, and
at last results in the formation of a large air space between
the peripheral cells and the base of the nucleus and inner
integument (cf. figs. 10, 14, 30, &c.)

About now, or a little later, the pollen-tubes appear in the
upper part of the ovary as silver threads, creeping slowly
down the tissue in the groove formed between the swollen
placentas and the carpellary walls. They may be few or
many, and in the latter case can be detected by the naked
eye on the walls of the ovary if torn open. Owing to the
prevalence of very cold and wet weather this summer I
cannot consider the question settled, but believe that the
further vigorous development of the embryo-sac and contents
depends on the presence of these tubes. Since insects were
rare during the bad weather I pollinated many flowers by
hand, and certainly got more vigorous ovules from the spikes
so treated, and there were many tubes in these ovaries. In
cases where the flowers had been pollinated naturally, how-
ever, the contents of the sac and the cells of the integument
were brown, and weakly developed. Selecting what appear
to be normal cases, the following changes occur in the
embryo-sac, which by its growth has considerably compressed
the layer of cells around it, and is capped by the remains of
the two upper cells (figs. 5, 6, 7, &c.).

The protoplasm collects into two masses, one at each end
of the embryo-sac, leaving the centre of the sac filled by

more fluid material;[1] for some time these masses are ill defined, and they never form a distinct cell-wall, but, finally, from the presence of a nucleus and well-marked contour, we recognise in them naked cells in a protoplasmic matrix (fig. 7).

Each of these soon divides into four smaller but otherwise similar nucleated masses, by two divisions in planes crossing at right angles. In some cases, at any rate, this occurs simultaneously in both, but in others one or both divide first into two, and then again each daughter mass into two others (cf. figs. 8—12).

That this is the final result there can be no doubt, but whether one is to suspect variations in the divisions as due to manipulation, conditions of vigour, or peculiarities of the species, must remain at present undecided. The important fact is that eight nucleated masses of protoplasm result, more or less isolated and complete, in groups of four at each end of the sac (figs. 12, 13); this I consider demonstrated by the specimen figured at 12, though the division of the lower mass does not always appear to be completed.[2] The relations of the products of division in the fore part of the sac are, however, remarkably constant and surprising. Two of them become elongated and packed close into the top of the embryo-sac, as the " Gehülfinnen " or " Synergidæ " of Strasburger; while one enlarges and rounds off as the egg-cell or " oosphere," and becomes suspended laterally at the base of the " Synergidæ " in the cavity of the sac. The fourth mass also rounds off, and falls freely into the lining protoplasm of the sac. All these masses have acquired nuclei, that of the egg-cell being especially large and bright, but no trace of a cell-wall appears around any of them.

As to the fate of the products of division in the posterior end, I can say very little positively. That four masses commence to form appears certain, but only in a few cases have they become completed; as a rule, I find a mass of protoplasm in this place with a variable number of nuclei in it, but in some cases (fig. 13) four masses occur.

It is to be regretted that this point has not been more successfully dealt with, and also that the exact origin of the

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[1] Many facts suggest that this division is of the same order as the two preceding divisions of the " embryo-sac mother-cell." If so, we must consider that cell as suffering division into four, the last division wall being a very weak and diffluent one.

[2] The lower group may apparently suffer less complete division in other plants also.

embryo-sac nucleus in Gymnadenia remains uncertain from my
drawings. According to Strasburger, the typical process is as
follows :[1]—The fourth nucleus from the group in the anterior
end of embryo-sac travels down, and meets one of the four
from the posterior end, and, fusing together, these two form
one large nucleus—the nucleus of the embryo-sac. The
three nuclei left behind are "antipodal cells" or "Gegen-
füsslerinnen." This process I have failed to observe
directly in Gymnadenia, but may remark in this connection
upon figs. 14, 15, 16, and 17, in each of which are indica-
tions possibly of some such process.

In the first case (fig. 14), we have an ill-defined mass of
protoplasm in the posterior end of the sac, and two large
well-rounded nuclei close by the egg-cell above ;[2] though
the bare possibility exists that one of these nuclei is on the
adjacent wall of an integument cell, since the case is not
isolated, yet it is offered here. In fig. 15 we have a large
bright nucleus below, and a faintly marked (badly preserved?)
body on the side wall of the sac. In fig. 16, both above and
below, appear good round nuclei. In fig. 17 the upper
nucleus has evidently travelled down, and now abuts upon
the mass of protoplasm at the base of the sac ; this mass is
well rounded, and presents four nuclei, or, more correctly,
two large nuclei, in a commencing stage of division.

Were it not for the fact that in Strasburger's drawings
and description this process is so definitely put, and also that
in other cases (esp. Ranunculus, Lobelia, Anthericum,
Butomus, and Alisma),[3] I have often seen two nuclei free
in the sac besides the antipodal cells and a normal egg appa-
ratus, the above evidence would not deserve to be so insisted
upon ; it is incomplete, and is little bettered by the sugges-
tion that in these highly specialised plants a process of
reduction has become still more reduced, and that even the
rudiments are unusually imperfect and uncertain. However
this may be, in *Gymnadenia conopsea* there are formed a
normal "egg-apparatus," a large nucleus of the embryo-sac,
and a group of "antipodal cells."

The pollen-tubes should now be somewhere in the neigh-
bourhood, and in fig. 17 we have an example showing the

[1] 'Ueber Befruchtung u. Zelltheilung,' p. 32, "*Orchis pallens.*" Com-
pare Vesque's different account of *Orchis galatea*, 'Ann. des Sc. Nat.,' 1878.

[2] Is it possible that this is a case of two egg-cells? Strasburger figures
two embryos in the same sac, in a paper published in Jen. Zeitschr. f. Wiss.,
1878—"Ueber Polyembryonie."

[3] In Butomus there can now be no doubt of the fusion of two nuclei.
I have every stage in the process. See a paper read before Linnæan Society,
Nov. 20th, 1879.

act of fertilisation; the pollen-tube, after a sinuous course from the placenta, has made a sharp bend ere plunging into the micropyle, and has then spread its broad apex over the " Gehülfinnen," apparently penetrating between the sac and integument, but the difficulty of tracing so delicate an out-line as it here presents is no ordinary one.[1] It does not break through the top of the sac, but the well-marked contour of the latter is to be seen through the transparent tube, the walls of which are here marked by delicate longi-tudinal striations, which appear to be folds, either caused by contraction from reagents, or the closing in of the inner integument; I cannot identify it with the filiform apparatus of Schacht, since it is on the pollen-tube, and has nothing whatever to do with the embryo-sac contents. In the course of the tube are developed the peculiar cellulose blocks pro-jecting inwards from its walls, and serving apparently to shut off the contents of the tube as it grows; these " Propfen " or stoppers were described by Strasburger[2] and Elfving,[3] and occur in such quantities that a cross section of the pollen-tube bundles appears marked here and there by waxy-looking drops interspersed among silvery-like cellular walls of the tubes. In the figure is one of these stoppers repre-sented as it occurred in the tube just ere the final bend.

The outline of the embryo-sac is still marked by the remains of the nucleus-cells which it compressed and des-troyed in its growth, but the cap-cells appear to have quite disappeared; even the latter, however, persist for a long time (cf. figs. 6—13) as a refractive cap on the apex of the sac, in some cases (fig. 12) presenting a conical, or even beaked appearance. If no pollen-tube enters the micropyle, the whole ovule turns brown, shrivels, and the contents of the sac become ill defined and decay; the egg-cell persists apparently longer than the Gehülfinnen (fig. 17, a). In all these cases, and up to a much later period, we find the remaining cells of the central or axial row persist beneath the sac; in fact, the air space already referred to is formed beneath the lower one, and between it and the outer integu-ment when the sharp bend is established.

The fact that the oosphere is fertilised is marked by the appearance of a thin cellulose coat around it; it elongates, and its nucleus prepares to divide. The contents of the Gehül-finnen and other parts of the sac, on the contrary, become

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[1] Hofmeister represents the tube as entering the sac in *Orchis morio.* ' Vergl. Unters.,' t. iv.

[2] Loc. cit.

[3] ' Jenaischer Zeitschr. für Naturwissensch,' 1878.

cloudy and prepare for degeneration, and final disappear.
Very shortly the remains of the Gehülfinnen form a lens-
shaped cap between the embryo and the apex of the sac,
while the decaying antipodal mass presents a similar appear-
ance below (cf. figs. 18—22).[1]   In fig. 21 the appearance of
four nuclei in the antipodal mass—and a similar condition of
things is vaguely indicated in other cases, as fig. 19—suggests
that the union of a fourth nucleus with one from above to
form the embyro-sac nucleus has not occurred, or if so, a
further division of the antipodal cells has taken place.

The nucleus of the elongated embryo-cell now divides, and
a wall appears between the new nuclei cutting it into an
upper and a lower cell.   This horizontal lamella is thin and
sharply marked, and is nearly or quite perpendicular to the
long axis of the embryo.   Each cell is full of fine grained
dark protoplasm, and its nucleus is very large, spherical,
and bright, and contains one or two brilliant nucleoli
(fig. 18.)   On account of the different fates of these two cells
we must distinguish them from the first; the upper one
becomes the pro-embryo or suspensor, and has but a tran-
sient existence; the lower produces the true embryo.

Each nucleus repeats the process of division exactly as
before, and we have the embryonic body divided by two
more walls parallel to that first formed (fig. 20), and already
is established a physical difference between the embryo and
pro-embryo, the latter being narrow and tapering somewhat,
and having diffluent thick walls, while the embryo rapidly
becomes stouter and more globular in accordance with the
distribution of its thin sharply marked cell-walls (cf. figures).
In fig. 19 the wall in the embryo is completed, and two
large nuclei again rounded off, but that in the pro-embryo is
only just appearing, its nucleus being fixed in the last stage
of division prior to the separation of the new nuclei, which
have already commenced to aggregate at the poles.   In
fig. 20 the process is completed.

A further difference between pro-embryo and embryo is
now established, in that the next divisions in the embryo
are perpendicular to that already formed, whereas in the pro-
embryo all the divisions are horizontal, and parallel to the
first.   In fig 24 the terminal cell of the pro-embryo is com-
mencing to divide as indicated by the condition of its nucleus,
while in fig. 27 the second cell has just divided, and the
last threads of protoplasm are still in contact with the new
wall.   We thus demonstrate that each cell of the pro-embryo
divides, and its elongation is effected by intercalary growth.

[1] In some endospermous ovules the antipodal cells divide vigorously.

In fig. 28 each of the four cells has doubled itself, and the power of division in the pro-embryo seems now to become exhausted, vacuoles begin to form, and the cells to elongate as their nuclei go to the walls, and the vacuoles collect into a sap-cavity (cf. figs. 28—30). Only in a few cases have I seen more than eight cells formed by the pro-embryo; in the specimen figured at fig. 30 are ten. As the cells elongate, since the solid embryo soon completely fills the embryo-sac, the apex of the pro-embryo becomes gradually pushed through the top of the sac, and the loose tissue of the micropyle allows it to escape into the cavity of the ovary (figs. 29, 30); its period of growth is now about completed, and, as the last divisions are made in the embryo, the pro-embryo turns brown and shrivels up, persisting as a mere ragged appendage in the ripe seed.

To return to the embryo proper consisting of two cells (fig. 20). It becomes broader, and the nucleus of one cell divides and a new wall cuts it into two equal cells arranged laterally. The plane of this new division is always perpendicular to that of the first (horizontal) wall, and passes through the longer axis of the whole embryo; it may appear first in the upper or in the lower cell, but usually the latter (fig. 21, 22). The other cell divides at the same time or very soon after by a wall, also passing through the long axis, and also perpendicular to the horizontal wall, but it is, in the majority of cases, if not always, also at right angles to the other perpendicular wall. Thus, in fig. 23 the first wall (horizontal) is cut at right angles by the second (perpendicular), which lies in the plane of the paper and in the upper cell; the third (perpendicular) will cut both of these at right angles or nearly so, as shown by the dividing nucleus in the lower cell. Similar relations are shown in figs. 24, 25. Each of the four cells thus formed rapidly becomes again divided by a wall perpendicular to all those which it cuts, and passing through the long axis of the embryo, and in this manner the embryo comes to consist of a nearly globular body cut into eight octants, in each of which is a large round nucleus (fig. 26).

The next division walls are again horizontal, and may appear first in the upper (fig. 29) or in the lower cells (figs. 27, 28); thus, the embryo becomes cut into twelve, and then sixteen cells, by walls in planes symmetrically related. A series of walls very soon mark out a central from an epidermal system; these (fig. 30) appear at about the same time in all the cells except the four which abut upon the pro-embryo, and lie in planes parallel to the outer wall of each. They may well be called tangential, and mark the first in-

dication of tissue differentiation; the cells of this outer layer only divide further by walls perpendicular to the outer surface. The ovoid embryo now begins to have starchy and other grandular matter deposited in its cells, and becomes thereby too opaque for observation of the cell-walls until submitted to the action of warm potash and glycerin or other clearing reagents. From the first no trace of vacuoles occurs, but the cells are tightly packed and full of fine-grained protoplasm, with large bright nuclei, and thin firm cell-walls surround them. By proper treatment, however, one recognises in the last stages of the embryo (fig. 31) that yet another series of tangential walls has appeared concentric to the first, and thus the ovoid mass presents a central column of cells, surrounded by a layer one cell thick, while over all is another layer, also one cell deep.

About this time the cells are crowded with nutritive matters, the pro-embryo and integument cells are empty and shrivelled (fig. 32), the remains of nuclei appearing on their dark brown walls, and the seed may be considered ripe.

In reviewing the processes above described, we may shortly point out several views held of late as to their meaning, and to render this more clear and complete, it may be advisable to recall to mind some points more fully dealt with in the larger text books.[1]

In the Ferns generally we have a spore developing a free chlorophyll-bearing, and often large prothallus, on which are produced antheridia and archegonia. When, as in Osmunda, the prothallus only bears antheridia at times, we may consider this the carrying to a step further a process common to this and many other genera, where the archegonia appear later than the very numerous antheridia. If we suppose the appearance of the archegonia indefinitely postponed the prothallus becomes unisexual—male; if prolonged after all the antheridia have decayed the prothallus is practically female in function.

Such a unisexal prothallus may be supposed diagrammatically represented in Fig. 1[1] as a section passing through the germinating spore, prothallus, and archegonium. In the long free neck of the latter are seen several masses of pro-

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[1] The English student has an excellent account in Sachs' 'Text Book.' Cf. also Hofmeister, 'Higher Cryptogamia,' Ray Soc., 1862, and Luerssen, 'Med. Pharm. Bot.,' B. i. Also literature quoted.

[2] The relative positions and sizes have not been insisted upon in the diagrams. In all the figs. *Ex.* = exospore, *En.* = endospore, *Arch.* points to neck of archegonium, and *Can.* to the canal-cells, *Oos.* = oosphere or egg-cell.

toplasmic substance (*can.*) which result from the partial division of a small piece of the oosphere, which becomes early cut off from that body ere it rounds off in the body of the archegonium ; these masses are the " canal cells " of the Germans, and by degradation become mucilaginous, and so serve to fix the antherozoids.
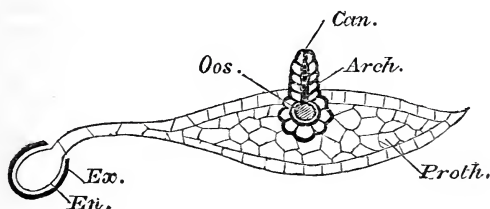
F<small>IG</small>. 1.—Diagram of free prothallium of fern, with its archegonium pro-jecting some distance exteriorly, and possessing a many-celled neck in the canal of which are the " canal cells."

Although the spores of Ferns present no external charac-teristics from which we can infer whether the prothallia to be produced on germination will be predominantly male or female, *i. e.* in its whole course of existence will bear arche-gonia or antheridia in excess, still we may see here indica-tions of a differentiation of function which, attended by reduction and abbreviation of the prothallus and processes peculiar to it, attains a limit in the highest plants. If in this specialisation and distribution we see an economy of material and energy, we can at the same time explain many of the phenomena.

In the Rhizocarps the separation of the sexes has been carried so far that from the spore itself we can predict whether archegonia or antheridia will be formed on its ger-mination, in some cases even the sporangia participating in the separation, and being distributed on different parts according as their products will yield male (microspore) or female (macrospore) prothallia. The male or antheridium-bearing prothallia are very small, becoming reduced to a mere tube of two or three cells in Salvinia, and in others being only represented by the trace of protoplasm left over from the antherozoid mother-cells. We must, in fact, look upon certain cell divisions in the microspore, preceding its germination, as representing the formation of a rudimentary prothallus and antheridia which form the few antherozoids then liberated.

In the macrospore, though the prothallus is also not set free, it is more obviously a cellular structure, producing one

or more archegonia. We may take that of *Marsilea* as a
type. The diagram (Fig. 2) represents a section through
the germinating macrospore, with its only partially exposed
prothallus, bearing an archegonium which differs from that
of the Fern in several points.
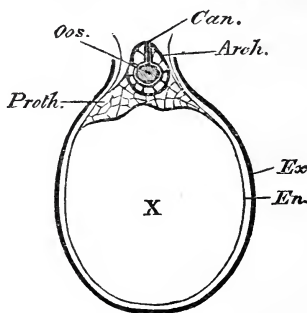
Fig. 2.—Diagram of partially free ♀ prothallium of *Marsilea*, it is re-
duced to little more than the archegonium, which hardly projects from
the surface, and has very few neck cells. The " canal-cell," however,
appears. The space ($\times$) becomes filled with fluid.

In the first place, its neck, instead of being a long, freely
projecting structure of several tiers of cells, hardly pro-
trudes at all, and is formed of two tiers of four cells each;
in plan these cells are arranged crosswise, and are almost
flush with the general level of the prothallium. Between
them, however, the young oosphere allows part of its sub-
stance, cut off as before, to penetrate as the " canal cell."

Besides a separation of the sexes, then, we have in Rhi-
zocarps a much smaller prothallus which never becomes
entirely free ; and as the prothallus tends to be withdrawn
(as it were) into the spore, so, too, the archegonia, &c.,
appear to be held back in the prothallus, and the neck to be
a less protruded structure. For a third stage in this re-
markable process we may select *Selaginella*. Here the
macrospores and microspores are not only produced in dif-
ferent sporangia, but the macro- and micro-sporangia are
borne on different leaves ; the sexes are further separated.

As before, the microspore undergoes the less extensive
development ; its contents becomes divided up into a few
cells, the majority of which produce antherozoids. The
spore then bursts and sets them free ; the process may be
considered as the formation of a rudimentary internal pro-
thallus reduced to little more than the antherozoid mother-
cells (antheridium). In the macrospore, a small prothallus
forms internally, and is just allowed to peep forth and

expose its one or two rudimentary archegonia. These consist of an oosphere, surmounted by four or eight neck-cells, which open flush with the surface, and have a " canal-cell " as before forced between them.
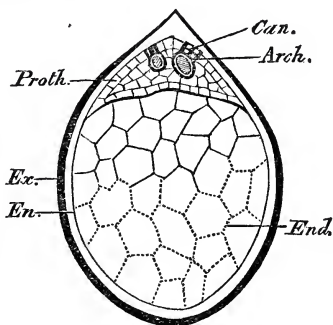
Fig. 3.—Diagram of endogenous prothallium of Selaginella, with few archegonia, reduced each to an oosphere with four or eight neck-cells as a " rosette " above, and with canal cell between. Before the rupture of the apex to the expose prothallus, a large-celled, delicate " endosperm " (end) forms below the prothallus, apparently independent of it.

These neck-cells form a kind of " rosette," as it has been termed, capping the oosphere which is sunk in the prothallus. But before the prothallus and its archegonia are exposed at the ruptured apex of the spore, a process occurs which results in the formation of an apparently new structure.

In Salvinia, Marsilea, &c., the space between the prothallus and the endospore, filling up the major part of the macrospore, becomes occupied by imbibed fluid, which accumulates and serves to push the prothallus upwards to the exterior, as it presses upon the " diaphragm " or membrane separating them. In Selaginella, however, a formation of large, thin-walled cells occurs in the fluid filling this space, and thus the so-called " endosperm " (Fig 3, end.) is produced. At a later period its cells become crowded with food material for the nourishment of the embryo, as this is pushed down by its growth and that of its " suspensor."

Our next step is to the Gymnosperms—the Conifers and their allies. Neglecting minor variations in detail, a typical Conifer presents the following features :

Its microspores[1] or pollen grains are produced not only in special and separate sporangia or pollen-sacs, but also on en-

[1] For further information as to these homologies, &c., the literature quoted may be consulted.

tirely different shoots, or even on other plants than those which bear the female sexual organs. Each pollen grain on germination (a process which by special appliances is brought about in close proximity to the female apparatus) emits a tubular body, in which indications of division occur, while a very few divisions are also established in the interior of the pollen grain itself. It is now generally accepted that the internal divisions represent a prothallium even more reduced than that of Selaginella, while the pollen-tube contents must be regarded as the representative of what becomes antherozoids in vascular Cryptogams—structures which are here rendered unnecessary as such, by arrangements already referred to. Instead of shedding motile antherozoids, therefore, the pollen grain carries its sexual products right into the region of the oosphere by means of the pollen-tube.

In the female apparatus we find the same principles of suppression carried still further; the macrospore[1] or "primary embryo-sac" is never shed at all, but germinates, so to speak, inside its sporangium—the so-called nucleus of the ovule.

During the gradual discovery of the phenomena which we are discussing, a number of synonyms have been introduced into the nomenclature, and some confusion is apt to arise in comparing these processes with what occur in Cryptogams; hence no apology is offered for the following summary:

The archegonia (the oospheres of which are the " corpuscula " of R. Brown, the " secondary embryo-sacs " of Henfrey) are formed by division of peripheral cells of a delicate prothallium formed of large, thin-walled, polygonal cells, and termed " endosperm ;" this prothallus arises in the protoplasm of a cavity which appears like an enlarged cell[2] of the nucleus of the ovule, and is the " primary embryo-sac." From its bearing archegonia and other relations, we must regard this " endosperm " as an internal prothallium similar to that which arises in Selaginella *before* the " *endosperm* " of that plant is formed; the term " endosperm " has been applied, therefore, to two structures which, whatever relations they may have morphologically, are differently distributed in time. The " endosperm " of Selaginella arises *after* the prothallus of that plant is formed, and coexists with it at the period of fertilisation; the " endosperm " of Conifers *is* the prothallus, and bears the archegonia.

The archegonium of Conifers consists of an oosphere, with

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[1] Or possibly several.

[2] And was so described by Hofmeister. But cf. Strasburger, 'Die Angiosp. u. Gymnosp.'

one or two tiers of four cells placed crosswise surmounting it; from the method of formation and the fact that in some genera a canal-cell has been observed cut off from the oosphere and forced up between these cells, this " rosette " may be
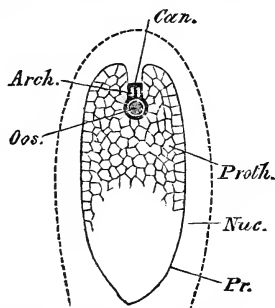
FIG. 4.—Diagram of wholly internal prothallium (so-called " endosperm ") of Conifer, with an archegonium, consisting of an oosphere (" corpusculum," " secondary embryo sac "), surmounted by a " rosette " of four cells, placed cross-wise; between these a " canal-cell " is forced. A tendency to still further withdrawal within the macrospore (primary embryo sac) is indicated by the funnel-like depression. *Pr.* Primary embryo-sac at apex. *Nuc.* Nucleus of ovule.

regarded as the neck of the archegonium. As seen in the diagram (Fig. 4) the archegonium is even more deeply withdrawn into the prothallus than was the case in Selaginella, and in some genera it becomes quite sunk into the prothallus.

We may now inquire what processes and structures in such an ovule as that of Gymnadenia are related to those just reviewed.

The first step appears to be to settle whether the " embryo-cell " of the Angiosperm is the equivalent of the " oosphere " of the Gymnosperm and Cryptogam.

Warming and Vesque argue somewhat as follows :—The " embryo-sac mother-cell " becomes divided by various transverse walls, just as the subepidermal cells of an anther become divided by walls parallel to the epidermis to form " pollen mother-cells." Hence, the cells into which the embryo-sac mother-cell is cut up are so many " spore mother-cells," *i.e.* each is equivalent to a pollen mother-cell. Vesque says further, that two of these opposed cells form each a group of four nuclei in its interior, just as the pollen mother-cell forms four pollen grains in its interior, and that the cell-wall between the two tetrahedral groups becomes absorbed, and so eight nuclei are formed, in two groups of four each, at the respective ends of the embryo-sac so formed by fusion of the two mother-cells.

They further point out that just as the cell-walls between the mother-cells of pollen are very deliquescent, so in the formation of the embryo-sac the dividing lamellæ are swollen and soon absorbed in similar manner. Also the four nuclei arising inside each cell (" pollen mother-cell" or constituent of embryo-sac) are arranged in tetrahedra in either case. If this account be accepted, the eight nuclei are homologous with pollen grains, *i.e.* spores, and the " egg-cell " is not to be regarded as the homologue of the " oosphere " of vascular Cryptogams, but as a macrospore which never germinates; and may be regarded as containing in itself the representative of the whole prothallus and archegonium.

But a careful survey of the facts in many different types has convinced Strasburger that no such fusion of two mother-cells occurs, and we see no such process in Gymnadenia as Vesque describes in *Orchis galatea* ;[1] especially does he appear to overlook the gradual compression and absorption of the two upper (cap) cells. The analogy between the mode of division in the embryo-sac mother-cell and the primitive cells of pollen-forming layers in an anther can probably be explained in a totally different manner,[2] and in any case we can lay no stress on it so long as it is unsupported by other evidence.

The following argument against this view appears to me important, especially if we take into consideration the many analogies tending to the conclusion that the Angiosperms are a series in which the reduction of the " prothallus generation " (oophore) is reaching its limit.

The pollen grain has been found to contain two nuclei,[3] apparently representing incipient germination changes—the first division in the microspore to form rudimentary prothallial structures. This being the case, from analogy with Cryptogams and Conifers (where we find it is always the microspore and male prothallus, &c., which suffer reduction first) we may argue the probability that whatever represents the macrospore will not have undergone more suppression than the microspore, and probably much less. But I think there is another reason for not imagining the female prothallus to be entirely atrophied, and for holding that the stages of reduction past a rudimentary archegonium are

[1] Loc. cit. Vesque's account of the facts also differs from that given here.

[2] See an attempt in paper to Linnæan Society, read Nov. 20th, 1879, "Contributions to Knowledge of Embryo-sac," &c.

[3] Strasburger, loc. cit., and Elfving in 'Jenaische Zeitschrift,' B. xiii. See the account of Elfving's researches in the present number of this journal.

almost inconceivable. For if the Synergidæ represent two cells of a prothallus, we may regard them possibly as remnants of the neck of an archegonium, and the egg-cell as its "central cell" (oosphere), and suppose that they have persisted in virtue of their use in reproduction.

Strasburger says that one cell resulting from the division of the embryo-sac mother-cell becomes the embryo-sac, that its contents divide as described into eight nuclei by three divisions in alternating planes; this embryo sac must be regarded as the macrospore—the equivalent of the embryo-sac of Gymnosperms.

Looked at thus, the divisions in the embryo-sac represent a rudimentary prothallus, and the "egg-cell" may be regarded as the oosphere. The "Synergidæ" were formerly thought to be the representatives of the "canal-cell;" but Strasburger, who at first took this view, afterwards showed that since they are sister cells of the egg-cell, and *not a product of its division*, such cannot be the case. He therefore considers them and the "antipodal cells," as well as the nuclei which fuse to form the "embryo-sac nucleus," as merely seven cells of the prothallus.

Strasburger, in criticising Vesque's theory, points out that no stress can be laid on the fact that the division walls in the embryo-sac mother-cell are deliquescent; for this, and the similar appearance in pollen-cells simply result from the imperfect nature of the process, since they will all be very soon absorbed, on the one hand by the enlarging embryo-sac, on the other by the pollen grains. The same remark applies to the divisions in the temporary pro-embryo of Gymnadenia.

Further, we cannot lay much stress on the tetrahedral arrangement of the nuclei, for the rule of rectangular division applies very widely,[1] and in the history of Gymnadenia we have seen how the first divisions even in the embryo follow a similar order; but in trichomes, prothalli, and other structures, the same obtains.

But another view appears possible. It will be remembered that the first divisions across the embryo-sac mother-cell follow one another in such a way that the two "cap-cells" were spoken of as being cut off from the mother-cell by diffluent swollen walls; and that, the lower cell having enlarged, destroying the cap-cells, its protoplasm passes to each end and a vacuole-like clear space forms between.

This last division may probably be looked upon as merely

[1] See also Sachs, in 'Wurzburg Arbeiten,' "Anordnung d. Zellen in jungsten Pflanzentheilen."

a third division across the embryo-sac mother-cell, and not as the first division of the contents of the macrospore (embryo-sac). In other words, we have here a division wall still weaker than the two preceding, and the " vacuole " is its expression. If this be so, it is possible that the embryo-sac mother-cell is really the mother-cell of four spores, two of which (the cap-cells) yield up their contents to their more vigorous neighbours—to the other two, which never completely separate, but form an " embryo-sac," and its contained apparatus.

This suggestion does not exclude the view that the eight nuclei derived from that of the embryo-sac mother-cell are cells of rudimentary prothalli, but explains them as belonging to *two* prothallial structures instead of one ; the one produces a rudimentary archegonium (the egg-cell with its synergidæ may perhaps be an oosphere and two neck-cells), and one vegetative cell ; the lower spore produces four vegetative cells. How to explain the subsequent fusion of one of these from each group is not easy, unless some advantage accrues to the large embryo-sac, by having a nucleus rendered vigorous by material from two slightly different sources. This is, however, too hypothetical to enlarge upon here, and perhaps we are yet far from possessing the facts necessary for an explanation of this remarkable process.

STUDIES *on the* POLLEN-BODIES *of the* ANGIOSPERMS.   By
FRED. ELFVING, of Helsingfors.[1]   With Plate IV.

UNTIL very recently botanists believed it to be a well-
established fact that the Pollen-bodies of the Angiosperms
were one-celled, that when once formed as tetrades in the
pollen mother-cell they underwent no further divisions.
This was thought, too, to form a direct contrast with the
Pollen-bodies of the Gymnosperms, in which, as is well
known, shortly before the period of pollination, one or more
so called vegetative cells are formed, which are regarded as
constituting a rudimentary male prothallus.

Strasburger[2] has, however, quite recently shown that the
Pollen-bodies of several Angiosperms, both Mono- as well as
Di-cotyledons, possess two nuclei ; he has further shown
that one of these originally pertained to a small peripherally-
formed cell, and that it only became free by the subsequent
resolution of the partition wall ; so that here, as in the case
of the Gymnosperms, a vegetative cell is formed in the Pollen-
body.

Strasburger further discovered in the case of the Orchideæ,
which he examined, that the nucleus of the large cell is
always in front in the Pollen-tube.

Strasburger points out that Reichenbach had already
figured and described both nuclei in the Pollen-bodies of
some Orchids ; and that Hartig had also rendered the two
nuclei visible in the Pollen-bodies of several plants by the
use of a carmine solution.   Yet these notices, because they
lay somewhat out of the beaten track, remained almost un-
observed.

I have attempted, at the request of Professor Strasburger,
to continue the researches into the stages of development of
the Pollen-bodies of the Angiosperms.   My researches, with
this view, were made in the summer time of 1878 in the
Botanical Institute at Jena, under the direction of Professor
Strasburger ; and this paper contains the result.   In these
researches I had the advantage of Professor Strasburger's
very kind and able help ; and I now gladly seize the oppor-
tunity of expressing my warmest thanks to him therefore.

My task consisted on the one hand in following up the

[1] Translated and condensed from the 'Jenaische Zeitschr.,' 1879,
part 1.

[2] "Ueber Befruchtung und Zelltheilung," 'Jenaische Zeitschrift für
Naturwissenschaft,' Bd. xi, Neue Folge Bd. iv, 1877, Heft. 4, p. 450.

newly-discovered cell division in the Pollen-bodies, and on the other in studying the relations of the nuclei in the Pollen-tube.

The reason why the fact of these Pollen-bodies having several cells has been overlooked by investigators, is probably altogether owing to the want of a suitable method of investigation. For if the phenomena which take place show themselves with surprising clearness in some plants, even without using particular reagents, yet, in general, little can be gained by the examination of fresh material; and the reagents formerly used by botanists rendered little or no assistance.

Osmic acid, which has lately come into use, proved to be an invaluable aid in these investigations. This acid has been used by Strasburger with great success in his investigations concerning the " Division and Fertilisation of Cells." After I had tried several other means for clarifying my specimens I used the osmic acid alone, and that only in a solution one per cent. strong. It is always of great advantage to add some kind of colouring material to the specimens preserved in osmic acid; sometimes it is quite indispensable to do so. A solution of carmine, to which a little glycerine was added, usefully served as such. By these means preparations can be obtained after a space of twenty-four hours which, in the way of clearness, leave nothing to be wished for. It is very useful to break up the Pollen-bodies, if they are large and richly filled with granular or oily contents, immediately after the osmic acid has been added; this can be done by pressing on the glass cover; for if left whole they may colour slowly or not at all. This is especially useful when it is desired to see everything quickly. The nuclei are in this way pressed out and immediately fixed, together with the rest of the contents of the cell. It is often even possible to get in this manner a view of nuclei in the act of dividing; colouring with carmine is naturally also here of great use. Plants with many-flowered inflorescences are especially suited for these investigations. If the first blossoms of such a plant have opened, it is easy to find all the younger stages of development in the buds above one another. The vegetative cell is, however, always (the case of the Cyperaceæ alone excepted) formed in those Pollen-bodies, which are separated from one another; that is to say, if they ever do separate.

In order to study the relations of the nuclei in the Pollen-bodies, I cultivated these in many and different solutions. This I did in the usual way—viz. in suspended drops in a

moist chamber.   At first I tried solutions of cane-sugar of
different degrees of concentration, but, as in many cases, no
satisfactory results could be obtained in this manner, I tried
other liquids.   Van Tieghem,[1] who also cultivated Pollen-
bodies, recommends the addition to the liquid of " une petite
quantité" of acid tartrate of ammonia.   I first of all decided
that this " petite quantité" could not be above 1 per cent., as
stronger solutions would be simply deadly.   Solutions of
·1, ·25, ·5, and 1· were, on the other hand, of no special use.
The directions again of Van Tieghem that inorganic salts,
sugar, gum, and etheric oils should be added, according to
the needs of the plants, are too general and uncertain to be
worth wasting time over in further trials.   After I had made
trial also of a solution of glycerine one per cent. strong, and
after trying solutions of nitrate of potassium and of carbonate
of soda, I turned back to my solutions of cane-sugar (called
in the following for the sake of brevity sugar solutions), as the
most suitable.   I made use of such solutions of different de-
grees of concentration (as 1, 3, 5, 10, 20, 30, and 40), for, as
might have been anticipated, it was soon apparent that the
maximum degree of concentration required is different for
the Pollen-bodies of different kinds of flowers ; while some
Pollen-bodies developed tubes in almost any solution, others
required a certain definite degree of concentration ; the indi-
vidual variations in this respect were very remarkable.   I
is of great importance to use, for the purpose of cultivation
quite ripe, though not over ripe, Pollen-bodies.

Omitting minor variations I have given in the following
pages the degree of concentration which proved itself the
most useful in the culture of the Pollen of the several
species of plants studied.   I have also given the time in
which the tube reached a certain length, under circumstances
which were otherwise normal.   Where no other solution is
mentioned the cane-sugar solution was the one used.

In all culture experiments the Pollen tubes swell after a
time into club-like bodies, and finally finish off by bursting.

In older tubes the still growing point is found separated
by peculiar cellulose-plugs from the emptied posterior por-
tions (see Strasburger, l. c., p. 456).

The culture experiments were undertaken with a tempera-
ture as warm as the usual one of a room and in the dark.
In many kinds of flowers the tube formation also took place,
and in a normal manner, in the daylight.   I did not turn my
attention further to this point.

At the same time I was, in the case of many plants, unable

[1] Annales des Sc. Nat., 5 série, t. xii, 1869, p. 318.

to obtain tubes artificially. I then, sometimes successfully, tried to make preparations thereof out of the already fertilised pistils.

The Pollen-tubes which I obtained in either way were immediately fixed with osmic acid or absolute alcohol. It is well to separate the moisture which surrounds the tube as much as possible before adding either of these reagents. This can easily be done with a capillary glass tube; and thus powerful diffusion streams are avoided, which often occasion the bursting of the Pollen-tubes. The fixing then takes place almost instantaneously.

The Orchids, especially those with richly-flowered spikes, offer splendid subjects for such investigations. I have examined *Orchis latifolia, O. mascula, O. maculata, Ophrys myodes, Platanthera bifolia, Gymnadenia conopsea,* and *Serapias francogallica,* and am able to confirm the statements of Strasburger in every particular; on which account I may for these simply refer to his drawings (l. c., p. 450, 452, plate t. xxvii, figs. 41—47). The formation and development of the Pollen-bodies corresponded exactly with these in every particular. The Pollen-body, originally provided with a round nucleus, is completely separated into two cells, of which the smaller is almost always to be found in one corner of the Pollen-body. The nuclei of these two sister-cells are round and almost of the same size; the body of the nucleus of the smaller cell is, however, always smaller than that of the larger. No trace of a cellulose membrane between the two sister-cells could be detected, either by the use of reagents or by the crushing of the Pollen-body. Even the separating plasma-layer is removed at a later stage, and in the ripe Pollen-body the two nuclei lie free beside one another.

The formation of the Pollen-tube takes a long time in the case of the Orchids, compared to that required by most other plants. Different species differ a little in this respect; the best results were obtained, as a rule, by a culture of from twenty to forty hours, and in a solution of from 5 to 10 per cent. of sugar. It proved to be, as Strasburger first found in these plants, that the nuclei wander together into the tube, and that in doing so the nucleus of the larger cell grows quite disproportionately. Both take a somewhat long elliptic shape. In tubes the results of older cultures, the anterior nucleus had elongated itself in a striking manner; the nucleolus was, however, to be seen clearly. I am inclined to regard this as only a change caused by cultivation; because in tubes extracted from styles one always

finds the nuclei of the same form, and this also when they are less developed.

I have tried to follow the nuclei until the moment of fertilisation. In *Gymnadenia conopsea*, used as a favorable object of research, I was able to make out definitely that the nuclei were to be found some distance from the point in those tubes which had already pressed their way into the micropyle, and whose point already touched the inner tegument of the ovule. In many cases one of the nuclei again divided itself; and so there were three nuclei. It always seemed to me to be the posterior nucleus which had divided, and in the case of *Orchis maculata* I can state positively that it was the posterior one. As soon as the fertilisation is over, which can be seen by the changed condition of the envelopes in the embryo sac, no further traces of the nuclei are to be found. The whole end of the tube, whose point lies on the embryo sac, often separated by a cellulose plug on the outside, is altogether homogeneous and highly refractive.

I may as well state here that I have brought ovules, which were capable of fertilisation, and tubes, when they (the tubes) were growing vigorously, together into one drop of saccharine solution; but that in no case could any entry of the tube into the micropyle be observed, not to speak of an act of fertilisation (Van Tieghem, l. c., p. 322, and Strasburger, l. c., p. 486). I will begin the description of the stages of development in the Monocotyledons which I examined, with the following, which I found to be one of the best examples.

Anthericum ramosum.—The Pollen-bodies of this species are almost semi-spherical. In a dry condition their convex side is deeply folded in. The extine is here very thin, and mostly broken through by the strong development of the intine, which later will evolve the tube.

In examining flower buds of about 5 mm. in height Pollen-bodies are generally found in which the vegetative cell is already formed (Plate IV, fig. 1). It is very difficult in the case of this plant to observe in detail the various stages of division on account of the dense contents of the cells. There is, however, no difficulty in ascertaining that the large circular nucleus places itself in the equatorial plain of the Pollen-body, and there divides itself. The result of the division is two cells. One, taking up by far the larger part of the Pollen-body, has a far larger nucleus, which is very like the original one, and like it has a very large nuclear body. The other, and far the smaller one, is attached to one side of the Pollen-body, and is separated from the sister-cell by a watch-

glass shaped wall, to which the intine is attached ; it is distinguished by its transparent almost homogeneous protoplasm, by its roundish oval nucleus, the nuclear body of which, though large, is still smaller than that of the larger cell. A partition by means of a cellulose membrane, as in the Orchids, seldom takes place, as the result of this division. The two cells are oftener only separated by a layer of cortical plasma, which originates from the " cell plate."

The vegetative cell soon separates itself from the intine altogether, and appears as a spherical formation in the inside of the Pollen-body (figs. 2 and 3). This cell elongates itself considerably in length and becomes spindle shaped with pointed ends, which are bent in (figs. 4—6) ; its nucleus remains almost unchanged. If the Pollen-bodies are carefully crushed in a saccharine solution (5 per cent. strong), this nucleus is easily found intact among the contents, which are pressed out. When pressed out of rather young Pollen-bodies, it generally becomes rounded and assumes a spherical shape, although it may have had already in the Pollen-body the spindle shape. In ripe Pollen-bodies, on the other hand, it is tolerably resistant, and keeps its form. Its nucleus appears clearer than the surrounding protoplasm if osmic acid is added ; it, however, stands out darker. When a strong solution of sugar—the best is one of 20 per cent.—is used, or, when a weak agent is added capable of absorbing water, the inner plasm structures will be seen to contract and to leave behind as a membrane the outer cortical plasm. The whole is coloured brown by a chloride of zinc solution.

While the vegetative cell changes in the manner just described, the nucleus of the big cell remains at first unaltered. It afterwards becomes longish, and in doing so often bends upon itself. Upon this its nucleolus also vanishes. It is very difficult to point out the nucleus in the ripe Pollen-body without using staining materials. The nucleus appears then as an irregularly-shaped, often crumpled body, or as membranous and shrunk together (fig. 6).

Anthericum liliago corresponds with the above species, except that the large nucleus keeps its round form. In a Pollen-body, which lay already on the stigma, but had produced no tubes, the nucleus was apparent, when fixed with osmic acid and coloured with carmine, as a strangely star-shaped body (fig. 8), and gave one the idea that it might have performed amœboid movements while it was being fixed. Direct observation on the living Pollen-bodies was impossible on account of the thick contents of their cells.

Pollen-tubes could not be obtained from these two species

by culture, I therefore prepared them independently from pollen-besprinkled pistils. In *Anthericum liliago* the whole vegetative cell is found in the tubes, together with the nucleus of the big cell, which is very much elongated and often looks like a very fine thread. It is usually this nucleus which goes first, though not without exception. Fig. 7 shows such a tube, which also exhibits the rather rare instance of branching. On the other hand, in *Anthericum ramosum* I could observe no nuclei as soon as the tubes were formed.

It is very much the same with *Globba bracteata*, only that in this case one can indicate no special place in which the vegetative cell will be formed, owing to the Pollen-body being spherical and provided with a membrane equally thick all over. In its ripe condition the Pollen-body is shaped like that of Anthericum (fig. 4). The Pollen-bodies, in a solution of 5 per cent. of sugar, produce short tubes; these, however, stop too short to allow of conclusions about the behaviour of the nuclei.

Tulipa Gesneriana (figs. 9—14) corresponds in most particulars with Anthericum. The vegetative cell is enormously developed (fig. 12 shows two pressed out), and its nucleus is often provided with several nucleoli. In the ripe Pollen-body it immediately strikes one from its size and half-moon shaped form. Occasionally I have found a division of the vegetative cell in the younger Pollen-bodies (fig. 13). In the tubes (1—3 per cent. sugar solution; eighteen hours) the nucleus of the big cell went before, and was followed by the elongated vegetative cell (fig. 14).

In *Ornithogalum pyramidale* the vegetative cell is also formed in a corner of the Pollen-body, opposite to the opening in the extine. In one instance, with osmic acid and carmine preparations, the division wall between the two cells clearly appeared double-contoured and was highly refractive like the intine, into which it undoubtedly passed over (fig. 15). I have no doubt that in this case a cellulose membrane was really formed.

Its further development corresponds with that which has already been described. The vegetative cell becomes finally much elongated by stretching, and has pointed, often bent in, ends. The greatest part of it is occupied by the almost cylindrical nucleus which possesses no nucleolus (figs. 16, 17). On crushing out the contents of a ripe Pollen-body the vegetative cell appears as a hyaline body in the midst. Both ends are filled with yellowish coloured little bodies; in general it has a strong resemblance to a cell nucleus, for

which it was taken in the like case of *Narcissus poeticus* by Strasburger. Osmic acid dissolves the yellow bodies, but brings out the nucleus clearly in the midst (figs. 18, 18*o*). The nucleus of the larger cell undergoes considerable changes, whereby the greatest part of its substance becomes dissolved, so that finally only a small, irregularly shaped, often whipcord-like remainder of it can be made out (figs. 16, 17).

Ornithogalum Ecklonii corresponds with the preceding. The Pollen-bodies in both species produced no tubes in the various solutions I used. There were no traces of nuclei to be seen in the tubes of *O. Ecklonii*, which I prepared from the styles, and which could be followed throughout their whole length; the ends of the tubes were thickly filled with fine granular protoplasm, and were cut off from the empty upper part and from the nucleus by the usual cellulose plugs.

Leucojum æstivum is a very favorable subject for examination, where the whole course of development, almost without using re-agents, can be pursued. The formation of the vegetative cell takes place in tolerably old buds. It is marked off by a thick arched-in wall of cortical plasm. The contents appears almost homogeneous (fig. 19); the addition of osmic acid causes, however, the nucleus, which possesses no nuclear body, to appear very distinctly (fig. 20). The question whether the vegetative cell is formed in a particular part of the Pollen-body I must leave unanswered, as I did not direct my attention to that point in examining it. Neither does my drawing allow me to come to any definite conclusion. The nucleus of the large cell is provided with distinct nucleoli.

The vegetative cell now loosens itself from the intine; at first spherical (fig. 21), it soon assumes the shape depicted in figs. 22 and 23. The largest part of the cell is occupied by the now elliptic nucleus, which appears pellucid in the fresh Pollen-bodies, while the protoplasm, permeated by dark-coloured granules, is pushed away almost entirely into the ends, which are bent in like horns. Fig. 23 shows such a vegetative cell after treatment with osmic acid. Fig. 24 shows the two nuclei crushed out and treated with osmic acid and carmine.

Shortly before the period of fertilisation, the wall of the vegetative cell is absorbed, and at the same time the nucleolus of the larger cell disappears, so that the two nuclei are hardly to be distinguished.

Pollen-tubes are to be obtained easily by cultivation (3 to

5 per cent. strong, six hours). The nuclei travelling into them are thereby elongated and become altogether similar to one another (fig. 25).

Narcissus poeticus.—Here the development is precisely similar, only that the vegetative cell is more spindle shaped, almost like that in Ornithogalum.

I obtained Pollen-tubes of *N. poeticus* after four hours' cultivation in a solution of 3—5 per cent. of sugar. In general, the nuclei, as soon as they have entered the tubes, cannot be distinguished, as they have become elongated early, often even in the Pollen-body (fig. 26). In many cases, however, viz. where the nucleolus of the nucleus belonging to the larger cell was still preserved, I was able to determine with certainty that sometimes the nucleus of the vegetative cell and sometimes that of the large cell went in front. I once saw, what is very exceptional, the doubling of the posterior nucleus (fig. 27).

Iris sibirica is very difficult of examination owing toe the richness of the cell contents and the thickness of th extine. The extine can be removed and the conditions of the investigation made easier if the Pollen-bodies are put into a drop of water or of sugar solution, and the covering glass is repeatedly raised up and down by means of a pair of small pincers. By such manipulation the extine will separate from several Pollen-bodies. If the Pollen-bodies of older buds are subjected to this treatment it is sometimes possible to press the whole contents of the large cell out, so as to leave the small vegetative cell attached to the intine (fig. 29). It must, therefore, be surrounded by a tolerably resisting (cellulose?) membrane, and I actually succeeded once in demonstrating such a membrane by a cautious crushing (fig. 28). The vegetative cell is always formed on the flattened side of the Pollen-body. Its nucleus has a small nucleolus, the nucleus of the larger cell is a little larger and has also a larger nucleolus. In the ripe Pollen-body the two nuclei are almost unchanged, the vegetative one is sometimes naked and sometimes covered with a hyaline protoplasmic mass of a spindle shape, which represents the vegetative cell set free.

Pollen-tubes were obtained after a cultivation of six hours in a solution, 30—40 per cent. strong, of sugar. As soon as the tube formation begins all signs by which one can distinguish the two nuclei vanish. They travel rather late, and often beside one another into the wide tubes, and can then only be pointed out as bits of protoplasm, coloured deeper than the rest by carmine, which have no clear outline.

Iris xiphium ; the development is similar to that of *I. sibirica.* I once observed one of its vegetative cells, whose nucleus had divided. The nucleus of the big cell sometimes goes first into the tubes and sometimes that of the small one. They also often go beside one another. The substance of the large nucleus becomes very much elongated on entering, and approaches the point of the tube by bending and twisting itself. The vegetative nucleus retains its shape even after the dissolution of its surrounding protoplasm (fig. 30).

Tradescantia virginica ; here the nuclei are very strangely formed (fig. 37), as Hartig has already remarked. One of them is very long and narrow, curved, with bent-in, almost rolled-in, ends ; it is, indeed, often of a trichina form. The other is round and finally of the same star-shaped form which is met with in *Anthericum liliago.* Both are devoid of nucleoli. The developmental history tells us that the former nucleus originates from the larger cell; while the other sausage-shaped one is the considerably modified vegetative nucleus. I may here mention in passing that the manner of division corresponded with that described by Strasburger for the integument-cells of *Nothoscordum fragrans* (' Ueber Befruchtung und Zelltheilung,' p. 517, Taf. xxxiii, figs. 47—54).

I examined the Pollen-tubes, which were prepared out of the style. These tubes grow out of one end of the Pollen-body. Already, whilst the two nuclei lie in the Pollen-body, the round one becomes much elongated, and after the two have entered the tube they are in every respect similar, becoming very long and drawn out. I discovered, however, some instances in which the vegetative nucleus lay in the Pollen-body with its characteristically rolled-up ends, while the other had already left it.

Sparganium ramosum shows a similar development to that of *Typha angustifolia.* The formation of starch (which hinders observation in Typha) begins, however, later, so that I can state positively that the vegetative cell generally separates itself and assumes a spindle shape (figs. 41, 42). The vegetative nucleus only becomes free after further development (fig. 43), after which its nucleolus vanishes (fig. 44). The nucleus becomes deeply coloured by carmine. The nucleolus of the large nucleus, deeper coloured, can generally be found in the ripe Pollen-body.

The nucleus of the large cell sometimes goes first into the Pollen-tubes (5—10 per cent., eighteen hours) and sometimes

the nucleus of the vegetative cell (figs. 45, 46). It, however, also happens that the latter may remain behind in the Pollen-body (fig. 48). The vegetative nucleus divides later on in the tube, and this occurs both when it is in front as well as when it is behind (figs. 45, 47). I never found it thus dividing while in the Pollen-body.

In *Pothos Olfersii* the two nuclei are similar also, being both elliptic and lying beside one another.

Not to mention the division of the vegetative nucleus in the Pollen-tubes of the Orchids and in Sparganium, I found that division of the vegetative nucleus may occur in *Iris xiphium*, and a duplication of the vegetative cell itself in the Pollen-body of *Tulipa gesneriana*. In these species this is exceptional. In some Monocotyledons such a formation of several vegetative cells is normal. This is well seen in *Andropogon campanus*. The Pollen-bodies of this species are spherical, with a thin extine which, as in all the Gramineæ, is pierced by a small orifice, through which later the intine grows out into a tube. In the stage of development in which the vegetative cell is formed, the Pollen-body contains only a thin parietal layer of finely granular protoplasm, surrounding a large vacuole. These Pollen-bodies are very favorable objects for examination, as starch and other bodies sometimes enclosed in protoplasm are altogether absent. Indeed, I have found no other plant which showed the formation and development of the vegetative cell so plainly, without making use of reagents. Pollen-bodies can be found in almost every anther; even in blossoms which are already ripe for pollination many Pollen-bodies appear in a retarded state.

Originally the Pollen-body carries only one single nucleus with a nucleolus. A small vegetative cell is then formed diametrically opposite the orifice in the extine. It is of the usual shape with clear protoplasm, and with a spherical or oval nucleus, which is provided with a small but distinct nucleolus. The nucleus of the large cell is generally disk-shaped, and has a large, strongly refractive nucleolus (figs. 54, 55). Typically, the vegetative cell next divides itself into two equal sister-cells, one of which often divides again, so that we have finally three vegetative cells (figs. 58, 59). The nucleus of the large cell remains unchanged. When this is over the protoplasm increases in volume and the Pollen-body fills itself with starch grains. Soon the vegetative cells are detached and float about freely (figs. 60, 61). Before the Pollen-bodies reach their ripe condition several nucleoli are absorbed, the nuclei themselves undergo

a lengthening out whereby the large nucleus visibly diminishes in volume.

Bromus erectus corresponds in many particulars with Andropogon. The original cell-nucleus has often as many as four nucleoli. The two daughter-cells have often more. Division takes place in this species tolerably late, as seems to be the case in most of the Gramineæ, in this case when the anther is about a centimeter in length. The vegetative cell, which is also here formed diametrically opposite to the orifice in the extine (figs. 61—63), divides itself, not immediately, as in the case of the last-named plant, but after the nucleolus is resorbed, first loosens itself away from the extine and now appears free in the surrounding plasm in which it sooner or later assumes a somewhat elongated form (figs. 64, 65). Then it becomes hidden by a copious mass of starch grains. I have only once seen the division into two of the vegetative nucleus in the uninjured Pollen-body (fig. 66), it is, on the other hand, very easy when the Pollen-bodies are placed in a 5 per cent. sugar solution, to find conditions like those in figure 67, where the vegetative cell has become divided into two still attached cells, forming a sickle-shaped body. The two vegetative nuclei are alike oval, and without any nucleolus. Later on they become considerably elongated, together with their surrounding cells, which are often found in the crushed-out contents hanging on by their ends, the nuclei being then often crumpled up; the nucleus of the larger cell with its larger nucleolus being still unaltered (fig. 68). At the last even this nucleolus becomes dissolved, whereupon the nucleus stretches and curves itself, so that finally it is not to be distinguished from the two vegetative nuclei, the surrounding protoplasm of which has meanwhile disappeared (fig. 69).

In *Butomus umbellatus*, as in all the Liliaceæ, the vegetative cell is formed directly opposite the opening in the extine, and somewhat late, namely, just before the buds are unfolded (fig. 75). The Pollen-body may remain in this state until the formation of the Pollen-tubes; a division of the vegetative nucleus into two generally takes place, however, either when it is still enclosed in the vegetative cell (fig. 76), or after the partition wall is absorbed. Lastly, these two vegetative nuclei are small and roundish, with an indistinct nucleolus; they are more deeply coloured by carmine than the nucleus of the large cell, which preserves its nucleolus a long time.

Tubes were formed, but rather sparsely, in a solution of sugar, 5 per cent. strong, after a space of twenty to thirty

hours, and I very often found tubes which had reached a con-
siderable length, and appeared otherwise quite normal, to
have within them the nucleus of the large cell, while the
vegetative cell, or vegetative cells, remained on the intine
quite intact (figs. 78, 79). In those cases in which the
nuclei of the vegetative cells were free they generally,
though not always, entered the tube, and that later than the
large nuclei (fig. 77).

Alisma plantago corresponds wholly with Bromus, except
that the place where the vegetative cell is formed cannot be
precisely indicated on the spherical Pollen-body. Attempts
at cultivation were fruitless.

In *Arum ternatum* the vegetative nucleus divides also into
two. This can easily be seen in Pollen-bodies, which are
crushed, and then immediately fixed with osmic acid (figs.
70—72). The two small vegetative nuclei are then often still
surrounded with their vegetative protoplasm (fig. 73). The
nucleus of the large cells appears wrinkled up in the fresh
Pollen-bodies; when it is subjected to pressure it assumes
the most strange forms, which are more or less star-shaped,
often even the "glove-shaped" form of Hartig (fig. 74).
Cultivation proved barren.

In the *Cyperaceæ* I found the most complicated phenomena
of development. I specially examined *Eleocharis palustris*.
Almost all stages of development can be found in the large
spikelets. At the time when the lowermost blossoms have
opened, the Pollen-bodies in the upper ones are still united
and polygonal; they have then a large nucleus in which
one almost always sees several nucleoli (fig. 82). Later
on the nucleus divides, and, indeed, it would seem is
complete usually before the separation of the individual
Pollen-bodies. As soon, however, as the Pollen-bodies have
become free and have taken their definite, almost spherical
shape, a division takes place of that nucleus which lies
towards the apex of the cell (fig. 84). One of the so
formed sister-cells divides again, so that we thus have
four nuclei in the Pollen-body, three small, oval ones
lying close to one another in the point of the Pollen-body,
and one large, more central (figs. 85, 86). In most
nucleoli, often several, are to be seen; that of the cen-
tral nucleus surpasses the rest in size. Only exceptionally
are four nuclei found in the point, giving a total of five.
The protoplasm, which fills the point and surrounds the
three little nuclei, often appears brighter than that in the
other part of the Pollen-body, and occasionally indications
of plasmatic partition walls can be seen, which mark the

boundaries of the nuclei; these, however, disappear soon, and the protoplasm then appears altogether homogeneous in the whole Pollen-body.

The central nucleus now divides (figs. 87—89). One of the sister nuclei is larger, and has a distinct nucleolus; the other is like those in the point, and, like these, has a small nucleolus. The three small nuclei are gradually absorbed after this division; they take colour in carmine solution, gradually less and less, and finally disappear altogether (figs. 90—92). As the two remaining nuclei often show several nucleoli, one might perhaps conjecture that here it is not a resorption of the smaller but a conjugation of the larger with the smaller nuclei that takes place, such as is the case in the embryo-sac. Observation, however, shows that several nucleoli may be already present before the small nuclei are absorbed.

Of the two nuclei which are now present, the one provided with the smaller nucleolus finally divides again (fig. 93); so that we have definitely three nuclei. One larger, with distinct nucleolus, and two small, which generally have either none or a very small nucleolus (fig. 94).

When these divisions have been accomplished the Pollen-body has become ripe. The extine has become clearly differentiated, and the contents include starch granules, and sometimes one finds ripe Pollen-bodies which appear to have two cells (fig. 98); closer examination, however, shows us that we have not here to do with a cell division in the usual sense of the word. The partition wall has rather been formed by an accidental fusion of peculiar inner thickenings of the intine, which is normally somewhat strongly developed in the apex of the Pollen-body (figs. 95 —97). In one instance I observed a nucleus in this so-formed chamber, which nucleus was clearly one of the three originally lying in the apex; its resorption had been prevented by the early formation of the partition wall.

The Pollen tubes (5—10 per cent., 5 hours) always grow either from the base or from the side of the Pollen-body, never from the apex. The nucleolus of the large nucleus vanishes at this time, so that it can only be distinguished by its size, and, in most instances, by its clearer colouring from the two others. All three suffer a diminution in size. The nuclei do not go into the tube in any particular order. In most cases, perhaps, it is the large nucleus which goes in front, and it is often elongated (figs. 99, 100). But other combinations may take place (figs. 101, 102).

Of the other Cyperaceæ examined *Carex vulpina* and *Cyperus badius* are, perhaps, less suited for examination than Heleocharis. I can, however, state with certainty that in all the development is precisely similar.

I have intentionally fully discussed the stages of development in the Monocotyledons. These can generally be more clearly seen in them than in the Dicotyledons, in which the richer contents of the cells and the small size of the nuclei make the examination more difficult. Besides, the Dicotyledons show nothing extraordinary; nothing which does not also occur in the Monocotyledons. I can, therefore, record my observations of them in a small space.

In the Pollen-bodies of the Dicotyledons we know that in general many orifices in the extine are formed. This, together with the more or less spherical shape of the Pollen-bodies, makes it impossible to indicate the place where the vegetative cell is formed with the exactness that one could do in the Monocotyledons. In the ellipsoid Pollen-bodies of the Papilionaceæ and Umbelliferæ, where the orifices for the tube are formed in an equatorial circle, the vegetative cell occupies a polar position. Even in the case of other Dicotyledons the position of the vegetative cell was never observed just under the extine orifices. Nevertheless, it seems to be the large cell that grows out to form the tube.

The vegetative cell is separated by a more or less convex partition wall of cortical plasm from the large cell. Its nucleus and nucleolus are always smaller than the same parts in the large cell.

The vegetative cell soon separates from the intine and appears as a spherical formation free in the Pollen-body.

As in many Monocotyledons, a division of the vegetative nucleus can still take place; so that three nuclei are present in the ripe Pollen-body. This occurs in *Sambucus racemosus, Fedia cornucopiæ, Dahlia Merckii, Nymphaea alba, Biscutella erigerifolia, Geranium Hookerianum, Arenaria laricifolia, Foeniculum officinale.*

The two vegetative cells resemble one another, and after a time have no nucleoli.

In *Nymphaea alba* the nucleus of the large cell generally preserves its nucleolus in the ripe Pollen-body, often even in the tubes (1—5 per cent., twenty hours). I can certify that this nucleus goes into the tube earlier than the two other vegetative ones which lie beside one another. It was only on very rare occasions that the opposite order obtained.

In the other plants mentioned above, the three nuclei can-

not be distinguished with certainty during the process of tube formation.

In most Dicotyledons no further divisions take place in the Pollen-body. In the following only two nuclei are found in the ripe Pollen-body, namely, *Gilia tricolor, Nicotiana tabacum, Salvia verticillata, Digitalis lanata, Gloxinia hybrida, Torenia asiatica, Plantago media, Campanula rapunculoides, Bryonia alba, Lysimachia punctata, Erica tetralix, Monotropa hypopitys, Peperomia claytonioides, Cannabis sativa, Rhus glabra, Ruta angustifolia, Ricinus communis, Hippuris vulgaris, Ranunculus muricatus, Delphinium decorum, Clematis viticella, Papaver dubium, Viola tricolor, Helianthemum polifolium, Ampelopsis hederacea, Oxalis lasyandra, Malva caroliniana, Polygonum rubrum, Begoniae sp., Sedum hybridum, Clarkia pulchella, Spirea villosa, Mimosa brachybotrya, Lathyrus silvestris.*

The development of all of these can be described in a very few words. The vegetative cell, which has become free, either remains spherical or becomes, as is more usual, spindle shaped. The parietal layer of cortical plasm sooner or later vanishes, and the two nuclei become mutually freed. These nuclei are, in their younger conditions, very easily distinguished. The nucleolus of that nucleus, which originates from the large cell, remains much longer than the vegetative one. This distinguishing sign vanishes, however; and in, or even earlier than, the tube-formation period, by successive metamorphoses both in form and size, the two nuclei become so like, that it is impossible to distinguish them. Only in *Cannabis sativa* is the nucleolus of the nucleus of the large cell preserved in the tubes (10 per cent., twelve hours). In this case sometimes one, sometimes another nucleus goes in front. In Monotropa the two nuclei preserve their nucleoli very long (5—30 per cent., twenty hours). An absolute distinction between them was nevertheless impossible.

Almost all possible differences occur in respect to the form of the nuclei, from the round one of Rhus to the greatly elongated, almost thread-shaped nucleus of Sedum. Generally they are almost elliptic in shape.

Putting together the chief results of my examinations it would appear that in a particular stage of development before fertilization the Pollen-body of the Angiosperms is divided into two cells—a larger and a smaller, the "vegetative" one—which latter, by further divisions, can form a two- to three-celled tissue (thallus).

This vegetative cell, or these vegetative cells are separated

as a group from the larger cell only by a wall of cortical plasm. In some isolated cases this may become a resisting (cellulose?) membrane.

The Pollen-tube is formed from the large cell. It may happen that the vegetative cell or cells have nothing to do with this occurrence, so that the nucleus and contents of the large cell alone immigrate into the tube. The separating wall is generally, however, absorbed. It may vanish altogether after the division; in most cases it however remains for a certain time; the whole vegetative cell or cells loosen themselves from the inner wall of the Pollen-body, and are then surrounded by the large cell, which appears strangely spindle- or half-moon shaped. The vegetative cell may remain in this state a longer or shorter time, or its nucleus may divide, and thus free-swimming vegetative cells be formed. In either case the wall of plasm is finally dissolved; this may take place either in the Pollen-body or after the vegetative cell has gone into the tube. A division of the naked vegetative nucleus may take place after the disappearance of the wall, and this may also occur either in the Pollen-body itself or in the tube.

The nuclei have often strange forms. I have noticed no division of the nuclei of the large cell, except in the Cyperaceæ.

A particular order is not generally kept in the migration into the tube. The nuclei are dissolved sooner or later; but in any case before fertilisation takes place. The large cell of the Pollen-body and its nucleus appear to be of more importance for fertilisation than the vegetative one. I arrive at this conclusion from the following facts, viz. that it is the large cell which grows into a Pollen-tube; from the circumstance that in some cases the nucleus of the large cell *always* goes in front, while I never saw the opposite case as a constant characteristic; that in those plants where this is not the case, the nucleus of the large cell still goes oftener in front than the other; finally, that in some instances the vegetative cell remains in its original position without entering the tube at all.

On the DEVELOPMENT *of the* CONCEPTACLE *in the* FUCACEÆ.
    By F. O. BOWER, B.A., Trinity College, Cambridge.
    With Plate V.

THE first careful notice of the conceptacle which I have
met with is that in the ' Phycologia Generalis ' of Kützing
(1843).

He describes (p. 98) it as a roofed-in sorus (einges-
tülpter sorus) ; while the tissue which lines the cavity is, he
says, nothing else than a slightly modified continuation of
the limiting tissue[1] (cortical schicht).

Speaking (p. 92) of the " Fasergrübchen " (a word which
has as yet no English equivalent), he says they seem to stand
in a certain relation to the conceptacle (Hüllenfrucht),
although they are found on such of the brown seaweeds
as have no conceptacles (cf. *Alaria esculenta*).

Agardh ('Species et ordines Algarum,' 1848, vol. i,
p. 101) suggests that the " Fasergrubchen " may be the
equivalent of the conceptacles (scaphidia) in the fertile part
of the plant.

Sachs ('Lehrbuch,' 1874, p. 283) remarks that the layer
of cells lining the cavity is a continuation of the outer limit-
ing layer of the Thallus.

The first attempt at an accurate description of the de-
velopment of the "Fasergrübchen" or conceptacle was made
by Reinke ('Bot. Zeit.,' 1875; and 'Nachrichten der K.
Ges. d. Wiss. zu Göttingen,' 1875, p. 230). His results
were republished in Pringsheim's 'Jahrbuch,' x, 1876,
p. 317.

Speaking of *Fucus vesiculosus*, he says (p. 337) that the
" Fasergrübchen," which he seems to take as the type of
these structures, originates, as seen from above, by a sepa-
rating of four or five neighbouring cells of the limiting tissue
from one another. He compares this process with the forma-
tion of the resin passages in the Coniferæ. The " intercel-
lular space " thus formed is filled with mucilage. Longitu-
dinal sections showed him that not only cells of the limiting
tissue, but also cells of the subadjacent cortical tissue, separate

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[1] Rostafinski ('Beiträge zur Kenntniss der Tange,' Heft i, 1876, p. 5)
has already objected to the use of the term " epidermis," in the case of
Fucus, on the ground of the outermost layer of cells being capable of con-
stant tangential division. He uses the term "aussenrinde," which I pro-
pose to render loosely by the term "limiting tissue," reserving the term
" cortical tissue," for his " innenrinde."

from one another.   An " intercellular canal" is thus formed, stretching through the limiting tissue into the cortical tissue. He further tells how the cavity thus formed becomes flask shaped, and the cells which line it put out papillæ, which further develop into hairs.   He assigns to the conceptacle a similar history, and hence concludes that it is " homolo- gous" with the " Fasergrübchen."   Though he records observations on a considerable range of allied plants, he finds no noteworthy deviation from the type of *Fucus vesciculosus*. He gives no figures illustrating these observations.

Luerssen ('Handbuch der Syst. Bot.,' 1879, Bd. i, p. 105) gives a different history.   He says small spots of the surface of the thallus are so overgrown by the surrounding tissue that only a narrow opening remains.

Thuret does not mention the subject of development of the conceptacle in any of his publications to which I have had access.

Preliminary Remarks.

Many of my preparations of *Fucus serratus* were made from materials collected in August, 1878; these were treated, while fresh, with a dilute solution of chromic acid in water, and afterwards preserved in alcohol.   The results obtained from these materials have been verified in other specimens, collected in August, 1879.   These were preserved in a satu- rated solution of common salt, and then hardened in alcohol. The latter method of preparation has been used for all the other plants of the group with which I have worked.   The sections were in all cases mounted in glycerine and acetic acid.

The youngest stages of development of the conceptacle are naturally to be found close to the apex of the branch, and only on those branches which still retain an active apical growth.   Such branches may be recognised by their possess- ing a well-marked depression at the apex.

Regarding the sexual conceptacle as the type of such structures, I have studied the development of it first, and then compared with it the development of the " Faser- grübchen," which I regard as an incomplete sexual conceptacle.

In describing the planes of section relative to the thallus, I use the terms " vertical longitudinal section " and " trans- verse section " in the same sense as Rostafinski ('Beiträge z. Kennt. d. Tange.,' Heft. i, 1876, p. 17), the former being a longitudinal section in a plane perpendicular to the flattened sides of the thallus : the latter cutting the organic axis at right angles.

Fucus serratus.

I chose *F. serratus* as the species best fitted for following the stages of development of the conceptacle for these reasons. (1) The fertile branches (Blüthen of Reinke) are less swollen than in other species, and it is therefore easier to obtain sections through the apex; while the tissues are more compact. (2) The hairs, which in later stages fill the cavity of the conceptacle, are not developed so early here as in other species. (3) *F. serratus* presents conceptacles of all ages in the month of August, at which time only I have had the opportunity of making collections.

The first traces of the conceptacle are best observed in vertical longitudinal sections.

In the fertile branches of *F. serratus* I have noticed no deviation from the form of the apical cells, and the succession of their segments, described by Rostafinski (loc. cit.) for the sterile branches of *F. vesciculosus.* The division of the lateral segments cut off from the apical cells also appears to follow the type of his fig. 14, each segment dividing first by a wall parallel to its free surface (" Basalwand "); the outer of the two cells thus formed again divides in two planes at right angles to one another and to the " Basalwand." Four cells are thus formed, each of which may again divide according to the same law as the original segment; this is the constant law of division of the cells of the normal limiting tissue of the younger branches, be they sterile or fertile (cf. fig. 1, groups marked *l*).

It is, however, in a modification of this succession of divisions that the first traces of the conceptacle make their appearance. Just as noticed by Rostafinsky (loc. cit., p. 9) for older branches, the division of the outer cell by vertical walls into four ceases in certain cases. The division by walls parallel to the surface (Basalwände), however, continues. A linear series of cells is thus formed which may be traced some distance into the tissues, but which is terminated by a single cell only (fig. 1). Later, the activity of division in the horizontal direction also ceases, and as the terminal cell of the series does not increase in size, the result is that it is surpassed by the tissues surrounding it.

The terminal cell of this series we may call the " initial "[1] cell of the conceptacle, the cell immediately beneath it may be termed the " basal " cell.

As the cells which abut laterally on the initial cell retain

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[1] Thereby conveying no functional connection with an apical cell.

the usual law of division, the result is that it is bounded laterally (as in fig. 1) not only by cells of the limiting tissue (aussenrinde), but partly also by cells of the subjacent cortical tissue (innenrinde).

Up to the stage represented in fig. 1 no important change is to be observed in the initial cell. It contains a nucleus and plentiful protoplasm, and its walls are normal. There may, however, be noticed at the apex the beginning of a separation of its cell-wall from those of the cells around it. In the latter the only modification of the usual divisions is an inclination of the vertical walls towards the initial cell.

Conceptacles of this stage of development are to be found on the inner side of the lips bordering the terminal cavity of the fertile branch.

Fig. 2 represents part of a vertical longitudinal section passing through a slightly older conceptacle. Here the initial cell has lost its internal tension. The cell-walls have collapsed, while the substance of the cell-walls themselves has been converted into the swollen mass which fills the cavity thus formed. This mass is continuous with the layer which covers the surface of the thallus; it already shows signs of the change which it undergoes later.

The upper part of the initial cell has shrivelled, and appears to have no cell contents; the lower part, which adjoins the basal cell, still contains a small quantity of vacuolated protoplasm. The basal cell has meanwhile increased in size, but has not yet divided. If figs. 1 and 2 are compared, it will not be difficult to assign their origin to the cells which border on the cavity in fig. 2.[1] It will be seen that, as Reinke rightly observed (loc. cit., p. 337), they are of different origin.

As foreshadowed in fig. 1, the cell-walls of the limiting tissue are strongly inclined in fig. 2 towards the centre of the conceptacle. The group marked l being most so, it is between the group l and the group c that the boundary of origin of the tissues lies, the group l being derived from the limiting tissue, the group c by the division of a cell of the cortical tissue. Those cells, then, which border the lower part of the cavity of the conceptacle (including the basal cell) are derived from the cortical tissue; those which line the upper part from the limiting tissue.

A surface-view of a conceptacle, intermediate in develop-

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[1] I do not mean to imply by such a comparison that the order of the cell divisions is constant. A mere glance at the figures will be sufficient to preclude such an idea.

ment between those of figs. 1 and 2, is represented in fig. 3. Among the normal cells of the limiting tissue, which appear thin walled and full of granular protoplasm, one is seen with contracted contents and swollen cell-wall. This is the initial cell.

The further development of the conceptacle as shown by section is marked by continued shrinking of the initial cell, and a gradual decomposition of the swollen substance which fills the cavity. This is accompanied by divisions in the basal and surrounding cells. In fig. 4 it will be noticed that the basal cell has divided in a plane parallel to the axis of the conceptacle. In the swollen mass which fills the cavity may be recognised irregular patches where the substance has undergone alteration, accompanied by change in optical properties. This is still more evident in fig. 5, which is part of a transverse section of the thallus.

Here the change of the substance filling the cavity has advanced so far that the unaltered portion immediately surrounding the remnants of the initial cell forms a central column of irregular outline. This stretches from the basal cell to the neck of the conceptacle, and is connected with the walls of the conceptacle by thin strings which, like itself, have remained as yet unchanged.[1]

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[1] We must draw a distinction between these two stuffs :—a, that which forms the central column and the layer which overlies the whole surface of the plant; and b, that which is formed at first in irregular patches, but which finally fills the greater part of the cavity of the conceptacle. The reactions, which are described later, bring to light a third body (c), which constitutes the outermost layers covering b at the exterior of the thallus (cf. reaction 6). The following observations will serve to point out their characteristics :

1. The substance a has different optical properties from b, so that they are easily distinguished under the microscope; a usually has a yellowish appearance.

2. The substance a does not change volume to any marked extent on dehydration; b, when dehydrated, contracts strongly, so that the conceptacle, when mounted in alcohol, appears only partially filled. When water is added the contracted mass swells.

3. Solution of iodine does not colour a, b, or c.

4. When treated with a solution of chromic acid in water, the substance b becomes more transparent, and swells strongly. a is only slightly swollen; and since its appearance is not much altered, it becomes more prominent, owing to the change in b.

5. With Schultz's solution neither a, b, nor c give a blue colour. b swells strongly; a is more resistant. [N.B. With this reagent the contents of the initial cell sometimes give the brown reaction of protoplasm; but this colouration is not constant, and probably depends upon the state of decomposition of the contents.]

6. Treated with concentrated sulphuric acid, b is immediately dissolved; a resists for a time, and is later dissolved; but a central portion remains

Besides the division shown in fig. 4, the basal cell at this time divides also in two planes at right angles to this and to one another, giving rise to a group of eight cells, four of which only can be seen at once in a section (fig. 5). These cells, during the further development of the conceptacle, appear to differ functionally in no respect from the other tissues bordering on the cavity. These tissues, having increased principally by divisions at right angles to the inner surface of the conceptacle, form a layer of thin walled, closely compressed, protoplasmic cells, which completely line the cavity; by these peculiarities this tissue is pretty sharply marked off from the adjacent cortical tissue.

Coincident with this increase in number of the constituents of the lining tissue is a change in form of the whole conceptacle. The lower part of the cavity becomes wider, so that the whole conceptacle assumes a flask form, which may be recognised in fig. 5, but becomes more pronounced at the stage represented in fig. 6.[1]

In the stage represented in fig. 6 the central column may often be found, still continuous, from the point, where the initial cell is fixed, to the neck of the conceptacle, which is, however, still closed. The column shows a jagged irregular outline, owing to the rupture of the strings which originally connected it (as in fig. 5) with the walls of the conceptacle. It may be noticed that the margin of the cavity is still unbroken by any outgrowth of hairs.

which is then seen to be continuous with the outer portion of the thickened covering of the limiting tissue, this we have designated c.

The conclusions to be drawn from these observations are, that a is a swollen form of cell-wall, which is not true cellulose, but is similar to the central portion of older cell-walls of the tissue of the larger tangles ("Innenlamelle," Luerssen, p. 101; " Gelin " (?), Kützing, ' Phyc. Gen.,' p. 34). That b is a substance coincident with mucilage ("Schleim," Kützing, ' Phyc. Gen.,' p. 30); that c is a substance akin to cuticle.

[1] It seems to me probable that the power of swelling with water possessed by the mucilaginous contents of the conceptacle, and the consequent internal pressure exercised on the internal surface of the conceptacle, has some connection with this form. Suppose such an internal pressure. The conceptacle is completely closed. If a yielding occurs it will naturally be at the point where the resistance is least, and that will be where the tissues are least crowded. This is, however, the case in the interior of the thallus. The cavity will then enlarge most at its base. Hence the flask shape.

This explanation is further supported by the fact that where the curvature is greatest (and the form of the conceptacle is usually less regular than in our fig. 6) there the cells of the lining tissue are largest. Hence it may be concluded, other things being equal, that there the external resistance was least. This supports our view. Again, it is universally the case that around the mouth of a conceptacle the surface of the thallus is raised so as to form a slight hillock (cf. figs. 6, 7). This would naturally be the result if an internal tension existed.

Not having the opportunity to make preparations from fresh materials, I was forced, in verifying these results, obtained with chromic acid materials, to use specimens preserved in saturated solution of common salt, and afterwards hardened in alcohol. Sections from these, mounted in glycerine and acetic acid, gave the same results in all important points; the distinction between the central column and the mucilaginous mass surrounding it being as distinct as in the chromic acid materials. When first mounted, however, in glycerine, the mucilage was marked by a very definite and beautiful system of striation and stratification; this was more striking towards the periphery of the mass, towards the centre the marking gradually lost its regularity. After the sections had lain in glycerine for a short time the appearance faded and disappeared, probably owing to a gradual swelling.

The direction of the systems is represented in fig. 7. In this figure it will be noticed that the outline of the cavity is irregular. This is due to the outgrowth of lumps of tissue, which, especially in male conceptacles, precede the formation of hairs.

Here we come upon the first evidence of a sexual difference. The conceptacles have thus far, in the diœcious plant, offered no variety in development according as they are male or female; but in the development and character of the hairs there is an appreciable difference. If the conceptacle be male the formation of hairs is preceded, as in fig. 7, by the outgrowth of irregular masses of tissue. The surface cells of these grow out into papillæ, which divide and form hairs, these hairs branch according to a monopodial racemose system; owing to their mode of origin these primary hairs are usually associated in bunches.

If the conceptacle be female the hairs arise more uniformly over the inner surface of the conceptacle, and are, as a rule, not branched. It is only after these hairs have begun to be formed that the initial cell and central column are thrown off. Up to this stage it may, in thick sections, be almost always recognised. Though the central column is detached, still the conceptacle remains closed at the neck for some time longer.

The antherida are not formed on the end of the upper branches of the primary hairs; the lower lateral branches of them are, however, at an early stage terminated by antheridial cells. Antheridia are formed also on the apex of small hairs developed for that special purpose by the outgrowth of cells of the tissue lining the cavity, and these show, in its

simplest form, the mode of branching which accompanies the formation of antheridia (fig. 8, i, ii, iii).

A single cell grows out to form a papilla. This divides (8, i) by a transverse wall. An apical antheridial cell is thus formed, which is supported by a single pedicel cell. The latter may now continue growth and put out a lateral papilla (8, ii), which is functionally equivalent to the original papilla.

In fig. 8, iii, a represents a system of antheridial branching of a higher order. $a\,i$ is an antheridium from which the antherozoids have already escaped, the primary axis was then $p—a\,i$. $a\,ii$ is an antheridium which was formed on the apex of a lateral outgrowth below $a\,i$. $a\,iii$ is the lateral papilla which precedes the formation of a third antheridium. The whole system is then, in this case, a sympodial system, of the type represented by Sachs in his 'Lehrbuch,' 1874, fig. 136, D (cf. 8, iii, B). It is only rarely that the branching is as simple as in the cases represented, since it is often not confined to one plane, and the antheridia are usually more crowded together (compare Thuret's figures, especially in his 'Études Phycologiques,' plates xv, 6 ; xxii, 8—10, xxiv, 6). Often the system approaches irregularly the type figured by Sachs (loc. cit., fig. c), but, however irregular, the antheridial branching seems always to be sympodial.

On the mode of formation of the oogonia I have no remarks to offer.

The difference between the " Fasergrübchen " and the true conceptacle is clearly marked in the mature state both by the form and the contents. But as we pass back to the earlier stages of development the distinction becomes less and less, till when we reach the condition in which the formation of hairs has begun in neither of them, it is impossible to say definitely, from mere inspection of the structure, whether the cavities will produce sexual organs or only hairs.

Still, if we take young plants and study the development of the cavities formed at their apex we may, with the greatest probability possible in such a case, regard such cavities as early stages of the " Fasergrübchen."

This I have done, and found their development to differ in no point of importance from that of the sexual conceptacle. The initial and basal cells are present ; the form of the cavity is the same. The central column appears just as in the sexual conceptacle.

The hairs themselves are multicellular but unbranched. The neck is usually wider than in the sexual conceptacle.

It does not open till the apices of the hairs reach the swollen mass, which closes it and push their way through it. The mass itself has been previously undergoing a process of disorganisation and swelling similar to that which takes place at an earlier period around the central column.

Seeing then that, i, in their development these two structures are so similar; ii, that " Fasergrübchen " do not occur among the sexual conceptacles on a fertile branch, *i. e.* the " metamorphosis " is complete; iii, that no function has been assigned to the " Fasergrübchen, except that they act physiologically as root hairs (Reinke, loc. cit., p. 321); iv, that they are completely absent from some plants, *e. g.* Halerica (Reinke, loc. cit., p. 360) and *Himanthalia lorea ;* I think the homology of the two structures is so clearly proved that I shall be justified in proposing instead of a translation of the German word " Fasergrübchen," we should use the name " neutral conceptacle " to denote these neutral structures. This may be objected to as a contradictory term, but I think it is important to convey at once the relation which appears to exist between them and [the true conceptacle. Since, however, the term conceptacle has now obtained a morphological rather than a physiological significance, the absence of the main function can hardly be taken as ground for refusing the term.

Fucus platycarpus.

The sexual conceptacles (*i. e.* those developed on the swollen branches) show in their earliest stages a structure corresponding closely with that of the sexual conceptacle of *F. serratus,* having an initial cell, terminating a basal series, which may, as in *F. serratus,* be traced into the substance of the thallus. In later stages, however, the initial cell appears in some cases to retain vitality and to divide, forming a central hair, of which the apical cell is partially decomposed and shrivelled ; in other cases it remains inactive, and decomposes as in *F. serratus.* The central column is not so clearly marked as in *F. serratus,* since it is very soon thrown off, owing to the early formation of hairs lining the cavity. For the same reason the identity of the initial cell is often lost at an earlier stage than in *F. serratus.* The division of the basal cell appears to be less definite and regular than in the former species.

In no point of importance were the neutral conceptacles found to differ from the sexual ; the hairs are formed earlier from the lining tissue than in the sexual conceptacles.

Fucus vesciculosus.

The conceptacles of *F. vesciculosus* resemble those of *F. platycarpus* rather than those of *F. serratus*, but the division of the initial cell is less frequent than in the former species. The development of the neutral is similar to that of the sexual conceptacles.

Ozothallia nodosa.

My materials, collected in August, 1879, supplied only the youngest stages of conceptacles. In these the initial cell and the divisions of the surrounding tissue correspond to those in the genus Fucus. The basal cell is, however, not so prominent as in *F. serratus*. In this point, and also in the general form of the conceptacle, Ozothallia appears to hold an intermediate position between Fucus and Himanthalia.

Halidrys siliquosa.

Viewed from above the young conceptacle of Halidrys appears in all respects similar to that of Fucus. In longitudinal section the initial cell appears in some cases as in Fucus ; but, in the large majority of cases, divisions occur in it, resulting at an early period in two or, in some cases, more than two cells. The basal cell divides at first by walls strongly inclined to one another. The cells thus produced do not at an early stage divide by walls parallel to the surface of the cavity. The result is that the conceptacle usually appears, as in fig. 9, as though lined by a layer of cells continuous with the limiting layer ; but as part, at least, of this tissue is derived from the basal cell, this conclusion is inadmissible. Meanwhile, the initial cell (or group of cells) has been completely thrown off by the swelling of the wall dividing it from the basal cell.

Later, as in the other plants of the group, the cells of the lining tissue put forth papillæ which develop further into hairs.

Himanthalia lorea.

Here we have a departure from the type of Fucus. Close to the apical cell of the fertile branch the cells of the limiting tissue, by means of repeated divisions parallel to the surface of the thallus, grow out into multicellular hairs, almost every cell bearing one. As the cells of this tissue continue to divide, some of the hairs are thrown off, and the rest remain scattered over the surface. These isolated hairs consist of an apical part, in which the cells are in various stages of decomposition ; a central part where the cells form

a beaded series; a basal part consisting of cells with only small quantity of protoplasm and slightly swollen cell-walls; this lower part of the hair gives the impression of having shrunk similarly to the initial cell of the conceptacle of Fucus. Tracing this hair back it may be seen, when viewed from the surface, to pass into a cavity between the adjoining cells of the limiting tissue (fig. 10). This cavity is formed, as in Fucus, by the shrinking of the lower cells of the hair and swelling of the contiguous cell-walls, so as to fill the space thus formed. Fig. 11 shows a conceptacle of Himanthalia at a slightly older stage in longitudinal section. The upper part of the hair has broken away, leaving only a single cell of the series behind. This cell is shrivelled and the cell-walls swollen, giving it the same appearance as the initial cell of Fucus, of which it is undoubtedly the equivalent. The form and size of this cell is very inconstant; sometimes two such cells of the initial series are left behind. There is no cell in Himanthalia which may be easily recognised as corresponding to the basal cell of Fucus. This is probably owing to the distortion of tissues which often occurs at a little distance from the apical cell.

During this change in the original hair, the cells of the limiting tissue, which immediately surround it, do not grow as rapidly as the adjacent tissue; they only divide rarely in a direction parallel to the surface; but more frequent and active division takes place in a direction perpendicular to it. The result is a depression of the surface around the initial cell. The tissue lining the cavity thus formed is therefore derived only from the limiting tissue; no part of it is derived from the cortical tissue. The further development of the conceptacle is carried on chiefly by divisions according to the same rule as at first. The region of greatest activity is at the base of the cavity. Thus the conceptacle attains the form of a long tube, which widens later at the base, so as to assume the usual flask form. As this widening proceeds, hairs are formed, as in Fucus, by outgrowth of single cells of the lining tissue. In a female conceptacle these hairs are multicellular but unbranched. In the male conceptacle the hairs are branched, at first on a monopodial system developed in a racemose manner; i. e. the apical cell divides transversely, and the upper cell grows on as before; the lower cell puts out a protuberance immediately below the dividing cell-wall. This does not, however, overtop the apical cell (cf. fig. 12). The apical cells of such hairs do not usually form antheridia; only the lower branches undergo the change. Coincident with the formation of the anthe-

ridia appears an alteration in the system of branching from monopodial to sympodial, as will be seen from fig. 12. Each of the lower lateral axes is terminated by an antheridium (1) which is cut off by a transverse cell-wall. This stops further growth in that direction. As before shown in the case of Fucus, below this cell-wall appears a lateral branch, which is in its turn again terminated by an antheridium (2), and so on. The system of branching is thus rendered sympodial by the formation of antheridia; the apparent axis being a pseudo-axis, composed of a series of pedicels of successive antheridia. As to the arrangement of the successive antheridial cells, the same remarks will apply here as were made upon the antheridia of *Fucus serratus.* As in Fucus, the antheridia are not only formed on the lower branches of primary hairs; unicellular papillæ are here also formed by outgrowth of single cells of the lining tissue; these develop as described in the case of *Fucus serratus* (cf. fig. 8, i, ii, iii).

General remarks.

In drawing our conclusions from the facts before us, the most striking point is that, in all cases described, *the formation of the conceptacle is preceded by the decay of one or more cells which occupy a central position with regard to the changes which follow.* The number of the cells thus removed is various, and the manner of their destruction is not constant; but the fact remains in all cases. I must own inability to suggest a parallel to this.[1]

A point which is not so obvious, but which appears of similar constancy, is *that the cell or cells which decay are in all cases members of a linear series.* It depends upon the activity of division, in a direction tangential to the surface of the thallus, how this series is characterised; whether, as in Fucus, where the division is slow, and even ceases, the apical cell of the series hangs behind the surrounding tissue; or whether, as in Himanthalia, where the division is often repeated, the series is elongated, and, protruding beyond the surface of the thallus, is called a hair. The small importance of this difference is shown by the variety which exists in this respect in specimens of the same species, under dif-

[1] Reinke ('Entw. Unters : über die Dictyotaceen,' 1878, p. 47) speaking of the hairs, which he termed "Sprossfäden," remarks that they are always unbranched; they are grouped in sheaves or rows, and grow, at least later, exclusively by division of their basal cells; they form the precursors of the reproductive cells, and fall off when these appear. How far these may be compared with the initial cell or hair of the Fucaceæ it remains for closer observation to decide.

ferent circumstances; e. g. the bunch of hairs at the apex of young Fucus plants is due to an unusual activity of tangential division in the cells of the limiting tissue. This phenomenon again appears at the apex of older branches of Fucus, whose activity of apical growth is in any way hindered (Reinke, loc. cit., p. 338). And Rostafinski (loc. cit. p. 9) notes how the cells of the limiting tissue of older branches divides by tangential rather than longitudinal walls. The activity of tangential division being then so variable, before asserting that the condition described in the case of Himanthalia is the normal one for that species, it will be necessary to compare the results here obtained with those drawn from materials collected at a different time of year. I think it quite possible that the plants I have used, being collected in August, were in a semi-dormant state previous to their period of active growth and reproduction in the winter. If so, the hair, as represented in fig. 10, may be compared with those at the apex of the young plant of Fucus, or of the older branch of Fucus living under difficulties. On this ground I shall not attempt to suggest which is the simplest form, or the typical mode of formation of the conceptacle.

This variation in the activity of tangential division accounts for the want of uniformity in number of the cells thrown off in different species, and even in the same species (this was especially noticed in Halidrys and F. platycarpus). In viewing the formation of the conceptacle of the whole group (as far as studied), we may say generally, that *the differences in mode of development* (in the early stages at least) *depend upon the difference in activity of tangential division of the cells of the central series;* for it will be seen that upon the behaviour of the central series in relation to the surrounding tissue depends also the origin of the lining tissue of the conceptacle.

In Fucus the terminal cell of the central series (initial cell), ceasing to divide tangentially and being left behind by the surrounding tissue (fig. 1), when that cell decays a cavity is formed, which extends further into the tissues than the base of the cells of the surrounding limiting tissue. The tissue lining the cavity is therefore in this case derived in its basal part from the cortical, in its upper part from the limiting tissue. In Himanthalia, however, the cavity thus formed only extends at most to the base of the cells of the limiting tissue; the layer lining the cavity is thus derived only from the limiting tissue. This distinction of origin must not, however, be put on a level with that from Der-

matogen or Periblem of the higher plants.   The mere fact that the cortical tissue of Fucus is derived by constant direct division from the limiting tissue is sufficient to show that the two are in the closest genetic connection, and that therefore the difference of origin of the lining tissue of the conceptacle is of very slight importance.

Upon the homology of the sterile conceptacle ("Faser-grübchen"), and of the sexual conceptacle, I need here offer no further remarks.

In comparing the neutral hairs in the male and female conceptacles of diœcious species, I find it the rule that *no branching occurs in the neutral hairs of the female conceptacle*.   It may be noticed in connection with this that no secondary lateral formation from the pedicel of the oogonium has been observed.   *In the male conceptacle even before the formation of antheridia the hairs branch*, they do so according to a monopodial racemose system.   It has been seen (cf. fig. 12) how the formation of antheridia modifies the branching to a sympodial system.   The antheridia were, however, observed to arise also on the apex of outgrowths from single cells bordering on the cavity of the conceptacle (fig. 8).   Comparing antheridia in the state represented by fig. 8, i, with the oogonia as represented by Thuret (Études Phycologiques, plate xii), we cannot fail to see a complete morphological identity.   Both are produced by the outgrowth of a cell bordering on the cavity of the conceptacle. In both cases a cell-wall tangential to the inner surface of the conceptacle divides an apical sexual cell from a basal neutral cell.   It is a matter of but minor importance that the basal cell of the antheridium may by lateral growth produce a second antheridial cell.   This may be regarded as an outcome of the general rule, that where the male organ is endowed with free motion, it is produced in greater profusion than the female organ.

We may conclude then that *the antheridium of the Fucus group is, at least in some cases, morphologically identical with the oogonium.*

Lastly, I must record my thanks to Professor de Bary, of Strasburg, for his kindness in allowing me the use of his laboratory, and also for his interest and suggestions during part of my work.

On CERTAIN EFFECTS *of* STARVATION *on* VEGETABLE *and* ANIMAL TISSUES.[1] By D. D. CUNNINGHAM, M.B., Surgeon, Indian Medical Service; Fellow of the Calcutta University.

THAT insufficient supply of nutritive material is closely connected with fatty degeneration of tissue is a well-ascertained pathological phenomenon. Rindfleisch, in his work on 'Pathological Histology,' expresses himself as follows in regard to this subject:—"This (the exclusively pathological category of fatty metamorphosis) includes all cases of disproportion between the means of nutrition and the parenchyma to be nourished. Such a disproportion may be caused either by a diminution of the *nutriens,* or by an increase of the *nutriendum.* If a minute vessel in the brain is plugged, the circulation in the area which it supplies is not wholly suspended, owing to the manifold anastomoses with neighbouring vessels ; nevertheless, a very considerable retardation of the current takes place, which may even give rise to temporary stasis and to hæmorrhage, and this suffices to disturb nutrition, and so to cause fatty metamorphosis."[2]

It is easy to find other concrete instances of fatty degeneration in association with impaired supply of nutritive material. The fatty degenerations of various tissues occurring in old age, in disused organs, and in tissues and organs where increase in bulk has been disproportionate to vascular supply, are currently cited as examples of it.

While this is the case, however, it is by no means generally recognised that fatty degeneration of tissue is an accompaniment of the general deprivation of nutritive supply incident on starvation. On the contrary, we find such a distinguished authority as Bauer writing as follows:—"During a period of thirty days of starvation the body loses the greater part of its organic albumen (*Organeiweiss*) without the cells becoming incapable of performing their functions; they continue to act and are capable of full recovery on the addition of material, and death results when the actions produced by the decompositions are no longer sufficient to permit of vital phenomena. There is

[1] This article forms a portion of a Report regarding the pathological changes observed by the author in persons who had died during the famine in the Madras Presidency in 1877. The complete paper appears as an Appendix to the recently published 'Fourteenth Annual Report of the Sanitary Commissioner with the Government of India.'—Editor ' Quart. Journ. Micro. Sc.']

[2] ' Manual of Pathological Histology,' by Dr. Eduard Rindfleisch (English translation), vol. i, p. 29.

here a gradual emaciation or atrophy of the organic albumen, but no breaking down of the cells into detritus."[1]

Although practical experience in regard to the effects of insufficient nourishment on the population of famine-stricken areas, as well as many phenomena in the organs of those dying of diseases incident on starvation, rendered the accuracy of such conclusions very questionable, it yet appeared necessary to investigate the subject experimentally, more especially as the interpretation of the phenomena presented by the tissues of subjects of famine dying of special diseases, such as diarrhœa and dysentery, is beset with certain fallacies dependent partly on the presence of the diseased conditions themselves as distinct from non-complicated starvation, and partly on the remedial treatment which may have been employed. The results of experimental inquiry into the subject occupy the earlier sections of this report as an introduction to the details regarding the phenomena observed in the human subject.

In regard to all the higher forms of life, whether animal or vegetable, it is now generally recognised that they are composed of living material, and of various structures developed from and by this material, but that these structures, whilst retaining more or less distinct structural evidence of their genetic relation to living material, cannot be regarded as themselves alive. Such are the walls of cell-cavities or of tubular or filamentous structures, &c., in animal and vegetable tissues. These are, no doubt, in many cases dependent for their integrity on the presence of the living material connected with them, but in others they may persist for various periods practically unchanged after the disappearance of the parent material, and in any case they have entirely ceased to exhibit any of the essential characteristics of living matter. They are never developed except under the influence of living material, but the latter can be developed independent of them, and can even persist and thrive for indefinite periods entirely apart from any of them. Changes occurring in such structures as the result of insufficient supply of nutritive material can only be induced mediately and through the direct influence of such deficiency on the living matter. The primary object of the experiments in the present instance was, therefore, the determination of the effects of insufficient nutritive supply on the living material, or, in other words, on the protoplasmic constituents of the tissues.

Professor Huxley long ago defined protoplasm as "the physical basis of life," and insisted on its general uniformity in character in whatever group of living beings it may be studied,

[1] "Der Stoffumsatz bei der Phosphorvergiftung," Von Dr. Jos. Bauer, 'Zeitschrift für Biologie,' Bd. vii, S. 63—85.

at the same time affirming that "it thus becomes evident that all living powers are cognate, and that all living forms are fundamentally of one character."[1] It cannot be supposed that any difference of opinion should exist on this point, and, such being the case, it must be evident to all that the effects produced on vegetable protoplasm by insufficient nutrition are of a nature analogous to those occurring under similar conditions in the protoplasm of animal tissues, and that the study of the former may afford much information in regard to the latter. Taking this into consideration, the reason that a series of experiments should have first been carried out on the influence of starvation on vegetable protoplasm becomes manifest.

In selecting particular vegetable organisms as the subject of experiment, the primary desiderata appeared to be—1st, to employ such as were capable of easy and accurate observation under the various conditions to which they were to be subjected; and, 2nd, to employ such as were familiar to the observer, so as to secure the best opportunities of fairly estimating the actual influence of various alterations in these conditions. Two species of mucorine fungi were accordingly selected. Both of these had been special objects of study for some years, and both are readily susceptible of artificial culture under conditions extremely favorable to accurate observation. An additional recommendation of these organisms as subjects of experiment lay in the fact that, due to their fungal nature, the processes of nutrition in them show a more accurate correspondence to those in most animals than they do in vegetable tissues containing chlorophyll.

In regard to animal tissues the great point appeared to be to secure such as could be submitted to observation during the life of the organism to which they belonged, or in any cases under circumstances involving the smallest possible amount of manipulative interference. In order to meet these requirements, the animals selected as subjects of experiment were tadpoles. From the extreme transparency of the tissues in certain parts of these organisms, and the ease with which large numbers of them may be procured and kept under observation, they afford exceptional facilities for the conduct of experiments; and although it may not be permissible to deduce exact conclusions on all points from the phenomena observed in cold-blooded animals regarding those which will present themselves in higher forms under similar conditions, yet in so far as the general effects of defective supply of nutritive material on the living elements of the tissues are concerned, there is good reason to suppose that the phenomena in both cases must be much alike.

[1] 'On the Physical Basis of Life,' Lay Sermons, Addresses, and Reviews, by T. H. Huxley, LL.D., F.R.S., pp. 120—146.

In regard to both animals and vegetables the procedure employed in carrying out the experiments was similar, consisting in retaining them for various periods in fluids in which nutritive material was present in various amounts, or almost entirely absent, the latter condition being secured by employing distilled water as a medium. With these preliminary observations a detailed account of the results of experiment may be entered upon.

I.—*Experiments on the Effects of Deficient Supply of Nutritive Material on Vegetable Tissues.*

The plants subjected to experiment were, as before mentioned, both members of the mucorine order of fungi; one of them being the sole known representative of the genus *Choanephora*, the other the well-known *Pilobolus crystallinus*. Both of these fungi are of constant occurrence in the neighbourhood of Calcutta, and both are readily susceptible of cultivation in media so liquid as to allow of the easy removal of germinating conidia and spores, or portions of mycelium and fructification, with a minimum of disturbance or injury. This is naturally a matter of moment when it becomes necessary to submit such structures to minute examination, or to transfer them to new media. In so far as Choanephora, however, is concerned, the nutritive fluid itself, a strong decoction of the corollæ of Hibiscus, is such as to allow of continuous observation of all the stages of growth in the fungal structures while still in it, and of ready dilution with, or entire substitution by, distilled water without any disturbance of the mycelium.

The conidia of Choanephora when introduced into the nutritive fluid germinate almost immediately, and under ordinary circumstances produce an abundant crop of conidial fructification within twenty-four hours. Immature conidia are full of a coarsely granular oily protoplasm, but they clear up greatly as they ripen, and ultimately the oily matter is only represented by aggregations of yellowish granules, one of which is generally situated toward either extremity of each oval conidium. When the conidia are subjected to conditions permitting of their germination, the first change which occurs in them is a gradual diffusion of the fat granules, and the establishment of streaming movement in the protoplasm. Ultimately the granules dissolve and almost entirely disappear, and the conidium, now full of shining, active protoplasm, emits a germinal tube. The latter grows and ramifies rapidly if the surrounding fluid contains sufficient nutritive material, and within the course of a few hours gives rise to an abundant mycelium. Where the fluid is deficient

in nutritive material, the mycelial development undergoes various modifications, and where nutritive material is absent, as in distilled water, many conidia refuse to germinate; and, in those cases where germination occurs, the germinal tubes are abortive, their growth being arrested so soon as the store material of the parent conidia is exhausted. The mere addition of moisture is sufficient to induce germination in many fungal spores and conidia, but where nutritive material is wanting, or where the means providing for its assimilation are absent, the amount of growth which takes place is determined by the amount of material stored up in the reproductive body. The phenomena observed in regard to the conidia of Choanephora afford an example of the result of absence of nutritive material, whilst those recorded regarding the germination of lichen-spores apart from algal elements appear to be referable to absence of assimilative capacity.

The germinal tubes and actively-growing young mycelial filaments of Choanephora are full of a shining protoplasm, in which sap vacuoles are present in varying numbers. When examined under high powers, the protoplasm presents a dimly clouded aspect, and a small number of brilliant, sharply-defined granules are seen scattered through it at wide intervals (*vide* fig. 1, A).
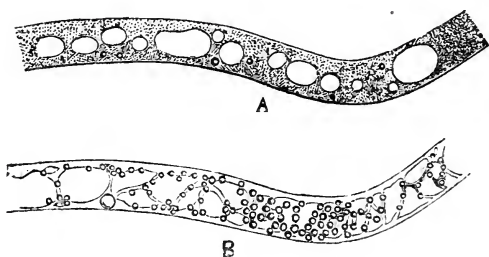
Fig. 1.—Germinal tubes of Choanephora.   × 1000.   A. Healthy tube.   B. Starved tube.

In such tubes it is almost impossible to detect any movement in the protoplasm, as, owing to the fulness of the tube and the general absence of distinct granules, there is nothing to serve as an index of movement save the slow changes in the form and distribution of the vacuoles. In older portions of mycelium, however, an abundance of granular matter accumulates, and here active streaming of the protoplasm is very distinct in many cases. Granular accumulation increases rapidly preparatory to the development of the fertile filaments, and these and the young

conidia arising from them are densely packed with coarsely granular matter.

This granular matter may be determined to be of an oily nature by the use of appropriate reagents, and its appearance in the fructifying mycelia and filaments of fungi is a phenomenon of frequent occurrence. It is usually ascribed to accumulation of oily matter derived from the nutritive medium, and to this, no doubt, it may in many cases be in great part due, but that it is entirely and invariably to be accounted for in this way is rendered very questionable by the phenomena which will presently be recorded as occurring in the protoplasm of starved mycelia. An excessive supply of nutritive material is certainly not favorable to the development of fructification, for where a portion of mycelium is retained in a concentrated and frequently renewed nutritive medium, the appearance of fertile filaments is indefinitely delayed, and the mycelium continues to grow luxuriantly, while a crop of reproductive bodies may generally be readily secured from it by diluting the medium or ceasing to renew it, and thereby allowing its nutritive properties to become exhausted.

Various other remarkable effects are produced by variations in the amount of nutritive supply, the special form assumed by the fructification in any cultivation being to a great extent directly determined by the quantity and quality of the nutritive medium. These will be referred to again in the descriptions of the effects of starvation of the mycelium, and a detailed account of them would occupy more space than can be devoted to it here. The general points to be borne in mind are—1st, that during the evolution of the young plants from the reproductive cells there is an involution and disappearance of oily matter; 2nd, that in young mycelia, in which vegetative growth is at a maximum and nutrition most abundant, the protoplasm contains a minimum of differentiated oily matter; 3rd, that in older mycelia, and especially in the fructifying filaments and young reproductive bodies, a great accumulation of oily matter occurs; 4th, that excessive nutrition appears to be antagonistic to the development of reproductive bodies.

Keeping these facts in view, the phenomena attending diminution, or deprivation of nutritive material, may now be described. The experiments regarding these may be referred to the following subdivisions : —1st, cultivation of conidia in distilled water; 2nd, cultivation of germinal filaments in distilled water; 3rd, cultivation of more or less developed mycelia in distilled water.

1st. *Cultivations of conidia in distilled water.*—In many cases, as previously mentioned, the conidia refused to germinate at all, while their contents became gradually resolved into a

mere loose aggregation of granular matter.   In other instances, however, germination occurred.   Evolution in such cases was limited to the development of short germinal tubes, and no formation of mycelium ever ensued.   The germinal tubes having been formed, two final results occurred.   These consisted either in a granular disintegration of the protoplasm, or in the formation of chlamydosporic reproductive bodies.   In the one case the contents of the tubes ultimately consisted of a collection of swarming oil granules ; in the other they became condensed into shining, isolated masses of oval or fusiform outline, consisting of dense aggregations of protoplasmic material, invested by a limiting layer, and in other respects resembling similar reproductive bodies occurring in other mucorine fungi.

2nd. *Cultivations of germinal tubes.*—In this case the conidia were introduced into nutritive fluid, and, at the close of some hours, when germination had freely taken place, the fluid was washed out and replaced by distilled water.   The healthy germinal tubes (*vide* fig. 1, A) contain an abundance of dimly clouded protoplasm, with numerous vacuoles and a few distinct granules. The primary effect of the substitution of water for the nutritive fluid is a great increase in vacuolation, the protoplasm becoming contracted as absorption progresses, and ultimately in the majority of tubes being reduced to an irregular peripheral layer, lining the walls of the tubes, and to a series of connecting processes extending across the cavity and forming irregular anastomoses with the lining layer.   Whilst these changes occur in the distribution of the protoplasm, another series of alterations affect its composition.   The number of granules present in it, which originally is very small, rapidly increases, and this increase combines with the altered arrangement in distribution to render the streaming motion of the protoplasm very evident as the granules are hurried along the peripheral layer and reticular processes.   The granules continue to increase in size and number, and assume a more or less distinctly oily aspect, and after the lapse of a few hours are very abundant and conspicuous.   After twelve hours of starvation all protoplasmic activity has ceased in most of the tubes, and the contents are now represented by a mere network of filaments studded with bright granules (fig. 1, B), but in some instances movement continues with great vigour, and is rendered particularly clear by the abundance and size of the granules.   Finally, however, movement ceases in these tubes also, and they come to present the same appearances as the others.   The peripheral layer of protoplasm appears to become gradually detached from the walls of the tube, so that the contents become reduced to a mere tangled aggregation of filaments and granules.   Eventually the connecting material disappears,

and the granules swarm free in the tube cavity, or unite into large oil globules, which may be dissolved out in ether, and are redeposited as globules and fat crystals on the evaporation of the reagent.

3rd. *Cultivations of mycelium in distilled water.*—The procedure followed in these experiments was similar to that in the case of those with the germinal tubes; the only difference lying in the fact that here the fungal elements were allowed to remain in the nutritive medium for a longer period, and until a considerable development of mycelium had occurred. As a rule, whilst the nutritive fluid was replaced by water in three or four hours from the period at which the conidia were sown in the experiments on the germinal tubes, the substitution was not carried out here until after the lapse of eight or ten hours. The primary effects of the treatment in the present series were identical with those described as occurring in the former one, but while in that series the process ran rapidly through various stages of fatty transformation and disintegration without the occurrence of any further development, one result of depriving a formed mycelium of nutritive material is almost invariably the development of fructification within the course of the next twelve hours.

The form of fructification characterising such cultivations is sporangial. As a result of previous experience, this form had been determined in the case of Choanephora to be developed under circumstances of defective nutrition dependent on various causes—on disproportion between the numbers of conidia sown in a nutritive fluid and its amount, on employing a fluid which was originally weak or had become exhausted by previous cultivations, &c.; but no other procedure appears to afford such certain production of a crop of sporangia. When the mycelial filaments have not been entirely emptied of their contents during this development, the condition of these is identical with that occurring in the case of germinal tubes which have been exposed to starvation for a similar period, and the development is probably to be ascribed to a utilisation by one portion of the protoplasm of materials derived from the disintegration of another portion.

In all three series of experiments the results were similar in showing that fatty change and ultimate disintegration of the protoplasm are direct results of insufficient nutrition. The development of reproductive bodies coincidently with these processes is very interesting, but by no means so anomalous as it might at first sight appear. There are many facts showing that the antagonism between individual growth and reproduction is not to be ascribed solely to expenditure in one direction counterbalancing that in another. Root-pruning and poor soil

will often force plants into flower and fruit, which, with a liberal supply of nutritive material, have persisted in mere continuous vegetable growth. This, however, is a subject hardly calling for detailed discussion here, but it is of importance to note that the coincidence of the formation of reproductive bodies with an oily condition of the protoplasm is of very frequent occurrence in fungi, and that the present experiments demonstrate that the presence of oil in such cases cannot be taken as necessarily indicative of excessive accumulation of nutritive material.

The other plants in which the effects of insufficient nutrition were studied—*Pilobolus crystallinus*—is in various respects not such a good subject for experiment. It cannot be satisfactorily cultivated in a medium allowing of continuous study without disturbance of the growing tissues ; it is impossible without great interference with and injury to the growing tissues to free them completely from extraneous matters from which they may derive nutrition ; and the healthy growing protoplasm in almost any case contains a considerable amount of differentiated oily matter. Allowing for these drawbacks, the experiments yielded results essentially similar to those in regard to Choanephora.

The materials for cultivation were furnished by the spores of the plant developed under natural conditions. The spores are at any time readily attainable in Calcutta, as portions of fresh cowdung kept for a day or two in a moist chamber hardly ever fail to produce an abundant crop of the fungus. Fresh spore masses, secured shortly after their discharge from the summits of the parent filaments, were introduced into cowdung, which had been diluted with water and subsequently boiled to secure the destruction of extraneous fungal elements. By this means a clean crop of Pilobolus was secured, developed in a basis sufficiently fluid to render the removal of germinating spores or mycelial filaments an easy matter. In removing mycelia it was found impossible to free the filaments entirely from adherent portions of the basis without injuring them so greatly as to complicate the results of experiment, but the amount of nutritive material was reduced to a minimum by the adoption of the following method. A mass of the cowdung containing mycelium was carefully removed, and introduced into a watch-glass containing distilled water. Gentle agitation served to wash out much of the cowdung ; the water was then poured off and replaced by a fresh supply, renewed agitation applied, and this process being repeated several times, the mycelium was at last obtained relatively free from adherent particles. It was then allowed to remain in distilled water, and the phenomena presented by it, at various intervals from the commencement of the experiment, carefully studied. As in the case of the other species, the main effect following the deprivation of

nutritive material seemed to lie in an oily transformation in the protoplasm, and the ultimate reduction of the contents of the filaments to mere accumulations of swarming fat granules. The nature of these granules can generally be readily determined by the action of appropriate reagents, and the quantities of yellow oil which can be extracted is very remarkable.

The spores of Pilobolus, as a rule, entirely refuse to germinate in distilled water, and the contents merely pass on into granular degeneration.

In these experiments on vegetable tissues the primary effect of starvation seems to consist in a gradual analysis of the complex amalgam constituting the protoplasm, and of a precipitation of oily matter from it. This is followed by transformation affecting the albuminoid constituents, and ultimately leading to their more or less complete conversion into fat. Vegetable protoplasm, according to most recent observations, consists of " a combination of albuminous substances with water and small quantities of incombustible material. In most cases it also contains, as may be concluded on physiological grounds, considerable quantities of other organic compounds, belonging probably to the series of carbo-hydrates and fats. These admixtures are distributed through its mass in an invisible form."[1]

The rapidity with which oil granules appear in many instances renders it probable that those first produced are the result of a separation of pre-existent oil. That the oily material subsequently produced is a new formation is demonstrated by the entire disappearance of all other elements from the interior of the cells or filaments.

The contrast between the phenomena attending evolution and involution is strikingly exhibited in these experiments. In germinating spores we find a solution and disappearance of differentiated oily matter ; in starved filaments, a precipitation of oily matter from a previously homogeneous protoplasm.

II.—Experiments on the Effects of Deficient Supply of Nutritive Material on Animal Tissues.

The animals employed in the experiments on this point were the larvæ of the common toad of this part of India (*Bufo melanostictus*) and of *Rana tigrina*, the so-called Bullfrog of Anglo-Indians. The procedure followed consisted in retaining the larvæ in water containing varying amounts of nutritive material, or, as far as possible, entirely devoid of it; freshly distilled water

[1] 'Text Book of Botany, Morphological and Physiological,' by Julius Sachs. Translated and annotated by A. W. Bennett and W. T. Thistleton Dyer. Oxford, 1875, p. 37.

being employed in the latter case, as in the experiments on starvation of fungal tissues. A very large number of careful observations were made on the toad-larvæ; those on the larvæ of the bullfrog were few in number, and only undertaken with a view to compare the general results with those of the former series. A detailed account will therefore be given regarding the phenomena observed in the toad-larvæ, with brief notices of those occurring in the others.

Before describing the effects produced by abnormal conditions of nutrition it is necessary that some account of the healthy tissues and organs should be given. A complete description of the entire anatomy of the larvæ would occupy much space, and seems unnecessary for a correct apprehension of the most important effects of starvation; anatomical details are therefore given in regard only to the transparent portions of the tails and to the intestinal canal. The state of the tissues in these was made the object of special study in all the experiments, the tissues of the fin affording exceptionally favorable opportunities for observations conducted during the life of the animals, and those of the intestinal canal being readily studied without the employment of means calculated to introduce manipulative fallacies.

The lateral portions of the tail in the larvæ of *Bufo melanostictus* may be described in general terms as consisting of a sheath of epidermis containing a network of ramified connective-tissue elements, together with vascular and nervous structures, and a number of free cells or bioplasts characterised by their irregular ramified outline and amœboid changes of form. A few cells containing pigment are also present in some cases, but their number and distribution are very uncertain.

The epidermis consists of two layers of epithelial cells. In the outer of these layers the constituent cells are flattened, so as to form polygonal plates. The edges of these plates are straight, or at utmost only show slight shallow sinuosities (*vide* fig. 2, A).

The cells vary greatly in size and form in various portions of the fin, but are apparently everywhere closely adapted to one another, forming a continuous, uninterrupted sheath over the deeper structures. Each cell contains a large oval nucleus, and this, together with the portion of the cell immediately surrounding it, appears to project on the under surface of the plate. Due to this circumstance, whilst the external surface of the entire layer of epithelium is flat and smooth, the under surface is uneven and covered with prominences. These prominences come into close contact with the upper surface of the inner layer of epidermis, while a series of shallow spaces is left elsewhere corresponding with the thin portions of the plates.

The inner layer of epidermis differs in many respects from that

just described. The cells of which it is mainly composed resemble those of the outer layer in their polygonal outlines,
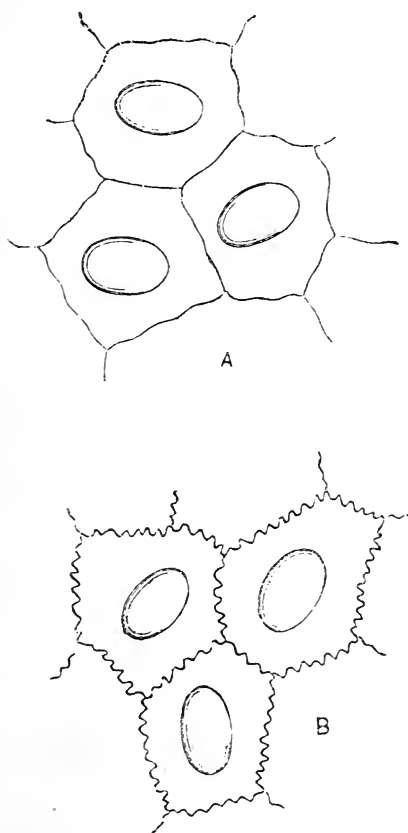
FIG. 2.—Epidermal cells. × 1000. A. Cells of superficial layer. B. Cells of deep layer.

and in the size and form of their nuclei. They are not, however, nearly so flat and thin, but present a tabular in place of a scale-like form, each cell being of some depth, and more or less convex inferiorly. The margins of the cells are deeply and closely sinuate, the prominences and depressions of neighbouring cells being closely adapted to one another (*vide* fig. 2, B). There is not the same uniformity throughout the whole extent of this layer of the epidermis as in the case of the outer layer. Intercalated among the cells which have just been described are cell spaces, which stand out conspicuously, due to their dark

granular contents, and, as a rule, considerably exceed the surrounding cells in size (*vide* fig. 3).
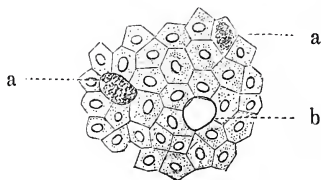
Fig. 3.—Deep layer of epidermis.   × 180.   a, a. Granule cells.
b. Empty cell-space.

The granules with which they are filled vary somewhat in size in different cases, in some being very coarse, and in others fine and molecular; when in mass they appear of a dark yellow or brown colour. In addition to these cells or cell spaces, for in many instances there seems to be no evidence of any distinct cell membrane, blank spaces or hiatuses in the continuity of the layer of cells present themselves here and there, which in size and form so exactly resemble the cells containing the granular matter as at once to suggest the idea that they owe their origin to the discharge of the contents from such structures.

The nature of the contents and the phenomena of development of these granule cells was carefully investigated, and I believe that there can be no doubt that they are merely epithelial cells which have undergone fatty change and which are ultimately removed to make room for new structures. At first sight they recall the " Schleimzellen " originally observed by Leydig in the integument of fish and amphibia, and subsequently described by various other authors, and specially by Franz Schultze and Langerhans.[1] Such cells are generally regarded as unicellular glands, which, according to some observers, discharge their secretion on the surface; according to others they never reach the surface, but by means of their thick mucoid secretion assist in effecting the desquamation of the older layers of epidermis. The granule cells in the present case resemble the " Schleimzellen " in being modified epithelial cells, but here the resemblance ceases. They differ entirely from the " Schleimzellen "

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[1] " Ueber die Haut einiger Süsswasserfische." Von Dr. Franz Leydig. ' Zeitschrift für wissenschaftliche Zoologie,' Bd. iii, S. 2, 1851. "Ueber Organe eines sechsten sinnes." Von Dr. Franz Leydig. ' Verhandlungen der Kaiserlichen Leopoldino-Carolinischen deutschen Akademie der Naturforscher,' Bd. xxxiv, Dresden, 1868. "Epithel und Drüsen-Zellen." Von Franz Eilhard Schultze. ' Archiv für Mikroskopische Anatomie,' Bd. iii, S. 137, 1867. "Ueber die Haut der Larve von Salamandra maculosa." Von Dr. Paul Langerhans. ' Archiv für Mikroskopische Anatomie,' Bd. ix S. 745.

in the nature of their contents; they cannot be regarded as unicellular glands unless all cells normally undergoing degeneration and destruction are to be regarded as such, and they certainly do not serve to facilitate desquamation, as they may be developed and ultimately disappear in excessive numbers without any tendency to desquamation.

The nature of the granular matter is easily determined by means of alcohol and ether, the granules being completely and rapidy dissolved under the influence of these reagents. The first effect of the solvents is to cause active swarming of the grauules, which either melt together into larger granules and globules previous to dissolving, or merely vanish, of a sudden, without the occurrence of such a process. The swarming granules occasionally become dispersed ere dissolving and spread for some distance in the interspaces between the two layers of epidermis; but I have never seen them discharged externally, even where the parent cells have been so much distended as to cause elevations of the surface. After the dissolution of the granules open spaces are left in the deeper layer of epidermis precisely similar to those occurring normally in it, save in cases where the cells have only undergone partial transfcrmation ere the application of the reagents, when traces of an atrophied cell-wall and nucleus are more or less distinctly recognisable in the spaces.

It is easy to trace the various stages of transition by which the normal epidermal cells are transformed into mature granule cells. The earliest symptom of impending transformation consists in an increased refractiveness and density of the cell contents. They acquire a yellowish tint, granules begin to appear, and, continuing to accumulate, from a dense yellow or brown mass, which ultimately occupies the entire cell cavity (*vide* fig. 4, A and B).
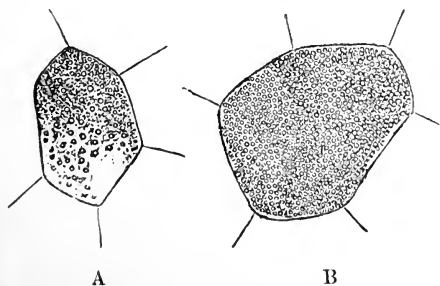

Fig. 4.—Stages in the development of granule cells. × 1000, A Young cell. B. Fully developed cell.

During this accumulation the cell generally increases considerably in size, so as to fill a space twice or three times as great as that which it originally occupied, and the nucleus is at the same time pressed to one or other side of the cell. After the highest stage of development has been attained, a certain amount of condensation usually occurs, and the granular mass shrinks and lies more or less free in the space which it formerly occupied. The nucleus and cell-wall finally disappear, and a mere mass of fat granules remains behind, lying in a large space in the epidermal tissue. Such appears to be the normal course of development of these structures, but in some cases more or less pigment is deposited along with the fat, and persists after the solution of the latter.

The only other bodies which these cells in some degree resemble are the granule cells discovered by Kölliker in the epidermis of the lamprey. They differ from them, however, in their simple structure, having no processes connected with them which can be regarded either as gland ducts (Kölliker) or sensory filaments (F. Schultze).

In the normal state of the healthy larvæ, the granule cells are developed in varying, but never in excessive numbers. In so far as a careful study of their development and subsequent history affords any information on their functions they appear to subserve important ends in the processes of growth of the tissue in which they occur, as well as in the involution of those portions of the latter which are peculiar to the larval condition, and disappear on the assumption of the adult form. As previously mentioned, the granule cells during the course of their development increase considerably in size, forcing apart the surrounding cells of the epidermis, and, on their dissolution, leaving spaces much larger than those occupied by the unaltered elements of the tissue. Under normal conditions these spaces are now filled up from below by the intercalation of new structures. These may in some cases be derived from the system of sub-epidermal free amœboid cells; but, as a rule, they are, I believe, derived from the connective-tissue system. I have frequently been able to trace a direct connection between fusiform nuclear bodies lying in the blank spaces and the common connective-tissue system of the interior of the fin; and in cases where this cannot be done, the appearances presented by the new elements in spaces which are about to be filled up cannot fail to render such an explanation of their origin probable. The peripheral ramifications of the connective tissue appear to be everywhere closely connected with the inner surface of the epidermis, a system of connective-tissue corpuscles lying scattered over the latter, while many processes appear to

be directly affixed to the epidermal cells themselves. On the formation of a blank space or hiatus in the epidermis, one or more connective-tissue corpuscles, according to the size of the area to be filled up, are drawn into it. The connective-tissue elements at first form an open reticular tissue, which, as the nuclei enlarge and the processes diminish, is gradually converted into a coherent patch of new cells intercalated among the older epidermal elements, and occupying a space originally formed by the degeneration and removal of one cell. If this interpretation of the phenomena be correct, one main function of the granule cells under ordinary circumstances is mechanically to enlarge the area of the inner layer of the epidermis.

Observations did not decide how the outer epidermal layer accommodates itself to the increased area of the inner one. The occurrence of any general desquamation was never observed, and there were no appearances of the addition of new elements from beneath. From the extreme variations in the form and size of the cells of the outer layer it seems not improbable that a good deal of the accommodation is accomplished by means of mere extension and flattening of the constituent cells.

It is evident that the granule cells may also play an impor tant part during the resorption and disappearance of the fin, for if developed in large numbers without equivalent addition of new elements from the deeper tissues, they must tend to the ultimate removal of the deep layer of epidermis. This, however, is a subject not calling for discussion on the present occasion. The important points to recollect here are, that when developed in due proportion, the granule cells provide for the growth of the tissue in which they occur, and that in such proportion their development is strictly physiological for the organism, although pathological for the individual cells.

The remaining structures in the tail of the larvæ call for little detailed notice, as their characters are familiar to all physiological observers. The branched amœboid cells beneath the epidermis are very inconspicuous during the life of the animal, and may readily escape notice unless carefully looked for (*vide* fig. 5, A).

The connective-tissue elements present no special peculiarities. The vascular loops are full, and the current of blood is strong and rapid. Many of the blood-corpuscles are characterised by containing two or three minute shining oil granules.

This brief account of the normal anatomy of the tissues may serve to render an account of the phenomena associated with deprivation of nutritive material intelligible. The following description is taken from the notes recorded during the examination of a tadpole which had been for a fortnight in distilled

water :—" The larva is very feeble and tends to turn over on its back when at rest. The thin portions of the tail to the naked
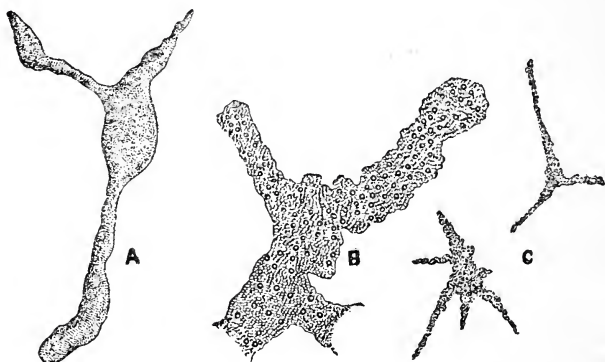
FIG. 5.—Amœboid cells. A & B × 1000, C × 180. A. Healthy cell.
B. Portion of a starved cell. C. Starved cells.

eye appear of a whitish colour in place of presenting the transparent aspect normal to them, and when examined with a simple lens are found to be covered with innumerable white points. Under higher powers these points are resolved into granule cells in various stages of development. Large numbers of fully-matured granule cells are everywhere present, and the number of partially developed ones is very great. Some of the latter are merely beginning to be distinguished from the surrounding epidermal cells by their shining yellowish appearance, while others present various degrees of granular accumulation. The tissue of the deep layer of epidermis appears, as it were, opened out, the constituent cells not being in such close contact with one another as they normally are. Empty granule-cell spaces abound, and here and there large areas occur in which the deeper epidermal tissue is either entirely absent, or replaced by loose reticular tissue consisting of nuclear elements connected together by slender processes (*vide* fig. 6).

" Granule cells sometimes occur, as it were, suspended in the midst of such reticular areas. The outer layer of epidermis appears little affected. There is no opening out of its texture, no hiatuses occur in it, the cells are everywhere closely in contact with one another, and the only abnormal appearances are presented by the nuclei, which in many cases are broken up into a collection of large shining granules. The branched sub-epidermal cells are greatly altered. They have lost their normal soft dim refraction and have become very conspicuous, appearing to

be distended with material of a yellow colour and containing
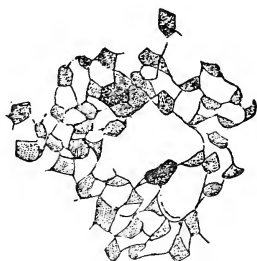
Fig. 6.—Reticular tissue replacing the deep layer of epidermis in a case of starvation.   × 180.

numerous refractive granules (fig. 5, B & C).   The connective-tissue corpuscles appear empty and shrunken, and together with their connecting filaments are in some cases studded with bright granules.   The amount of blood in the vessels is very small, the current is very feeble, and the corpuscles appear to escape from the vessels with abnormal readiness.   Many vascular loops remain entirely empty for considerable periods at a time, and in others isolated corpuscles slowly follow one another at wide intervals.   Almost all the corpuscles are studded with large granules and globules of oil, and many of them are quite bleached and colourless.   Some in process of distintegration can only be recognised by means of the characteristic arrangement of the oil globules which they contain, and free oil globules afford evidence of the complete destruction of others.   Alcohol and ether produced their ordinary effects on the granule cells, and prolonged application of the same reagents showed that the yellow matter and granules with which the sub-epidermal cells were filled was also of an oily nature.   The granules first vanished, and the contents of the cells then gradually escaped as large yellow globules, which gradually dissolved and disappeared.   The coarse granules in the nuclei of the outer layer of epidermis seemed to be little, if at all, affected by the reagents."

Numerous careful examinations of specimens in all stages of starvation afforded results substantially in accordance with those just recorded, variations in degree of change being naturally present in accordance with the varying periods of starvation. The phenomenon which presented itself most conspicuously and frequently at first was an increase in the number of the granule cells.   This increase cannot be ascribed to mere accumulation of the cells formed in normal proportion, but failing to undergo complete transformation, for—1st, the increase occurs

without any evidence of any equivalent increase in persistence of the cells, blank spaces being present in increased rather than diminished quantity; 2nd, the increase in number of granule cells is often extremely marked when there has been a very considerable destruction of the epidermal tissue over wide areas. In proportion to the period of starvation there is a steady increase in the number of blanks in the deeper layer of epidermis and of areas occupied by reticular tissue. This tissue, which in areas formed early in the course of starvation, is comparatively close and approaches new epithelial tissue more or less nearly in its characters, alters in nature in the later stages. The nuclear bodies are then small and far apart, and the connecting processes proportionately lengthened. Ultimately, in the last stages of inanition, the tissue in many places fails to be developed at all, and large blank spaces are left, in which the internal tissues are only covered by the outer epidermal layer.

The increased formation of granule cells is soon and constantly accompanied by changes in the blood supply. The amount of blood becomes visibly diminished, and the blood current flows with diminished rapidity. The oil granules in the corpuscles increase in size and numbers, so as to present themselves very conspicuously even to low-power observations. The destruction of the corpuscles, however, goes on very gradually until a late stage of starvation, at which it appears to assume a more active course. The colouring matter escapes from the corpuscles, and the latter ultimately break up, setting their contained oil globules free in the serum (*vide* fig. 7).
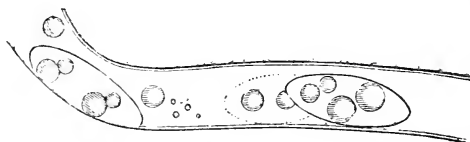
Fig. 7.—State of the blood-corpuscles in advanced starvation.   × 1000.

Somewhat less constant than either of the above-described phenomena, but yet of very frequent occurrence, is a fatty change in the ramified sub-epidermal, amœboid cells. They increase greatly in size, and appear to become distended with yellow matter and numerous granules. These changes are so conspicuous in many cases as to render the cells one of the most striking features in the starved tissues. The contents of the cells are dissolved out by ether, leaving mere shrunken skeletons behind.

No such conspicuous changes affect the connective-tissue elements even in very advanced stages of starvation. The nuclei

lose their plump, shining aspect, and numerous fat granules frequently present themselves, both in them and in the connecting processes, but no great accumulation of fatty matter or destructive change seems to occur.

The principal effect of defective nutritive supply on the tissues in these experiments, as in those in regard to vegetable tissues, seemed to lie in a fatty change. This change or degeneration specially affected epithelial elements containing a large proportion of living material, and the blood cells. The destructive changes in the blood cells must be regarded as a direct effect of defective nutrition, but it is not easy to determine how far the changes in the epithelial tissues are to be ascribed to this and how far to defective oxidation and consequent accumulation dependent on the changes in the blood. The persistence of the outer epidermal layer is very remarkable, and is probably to be ascribed to the extent to which it is composed of formed, more or less cuticularised material.

The normal anatomy of the intestinal canal and the changes occurring in it during starvation remain now to be described. The intestinal canal of the larvæ is of such simple structure and such transparent texture as greatly to facilitate the determination of these. All the structures can be readily subjected to observation without disturbing their normal relations, or interfering with the intestine in any way beyond removing it from the body of the animal. In normal healthy specimens the spiral coil of the intestine appears conspicuous externally, shining through the transparent abdominal walls. It consists of three coats, the external of which is muscular, the internal epithelial, and the intermediate one composed of reticulate adenoid tissue (*vide* fig. 8).
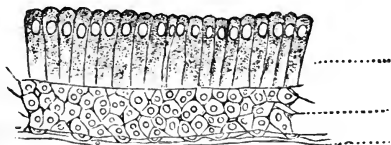
Fig. 8.—Normal structure of the coats of the intestine.   × 180.

The muscular coat consists of the two layers of fibres, those in the outer layer running parallel to the length of the canal, those in the inner transversely arranged. The epithelial coat is composed of large cylindrical cells. These call for no special note, as they closely resemble those occurring in the intestines of other animals. Their contents consist of soft, clouded protoplasm with a few scattered granules, and their only peculiarity lies in the fact that

their inner free extremities do not present such a distinctly defined border as that apparent in many intestinal epithelia. The reticular layer between the other coats is formed of a network of fine filaments, the meshes of which are occupied by free nuclear bodies. It is very thin, and under normal circumstances is very inconspicuous, lying, as it were, compressed between the other two coats. When the latter, however, are separated from one another by pressure, or by means of reagents which cause the inner one to contract, the meshes of the reticular tissue are stretched and open out, showing that in many places several strata of reticula are present. In some places, on the other hand, there appears to be only a single layer, the bands of tissue seeming to pass direct from the inner surface of the muscular coat to the outer surface of the epithelial one. Fig. 8 represents a specimen in which several strata of reticuli and nuclei are present. The nuclei are plump and shining, and after treatment with alcohol appear softly molecular.

The appearances presented by the intestinal canal of a larva in the last stage of starvation were the following :—" The intestine cannot be detected from the exterior of the abdomen, and on its removal from the body it is seen to be very slender and short. The canal is quite empty throughout almost its entire length, but here and there an isolated brownish-yellow granular mass is present. The epithelial coat is everywhere entirely wanting. The reticula of the adenoid coat appear not to have been in any way affected, but the nuclear elements present most abnormal appearances. In place of being quite colourless and smooth, they are rendered extremely conspicuous by being of various shades of yellow and brown, due to the presence of accumulations of granular matter within them. In some places comparatively few nuclei are present, and the reticula are empty or contain collections of free granules. The changes in the adenoid tissue are most marked in the upper portion of the intestinal canal, and in the large intestine a large number of unaltered nuclei are still present. On treating the specimen with alcohol and ether, active swarming of the granules in the reticular meshes and in the nuclei occurred ; and on continuing the application of ether, many of the nuclei and nuclear masses were entirely broken up, all the yellow matter being dissolved and disappearing, while an abundance of deep brown pigment granules remained behind. Many of the pigment granules lay loose in the reticular spaces, but others were contained within nuclei, or formed dense aggregations corresponding apparently with nuclei which they had replaced. The yellow granular matter was also dissolved out from the masses of intestinal con-

tents, and the oil thus extracted was subsequently precipitated in the form of abundant oil globules and crystals."

The phenomena presented in this case were not peculiar to it, but may be taken as fairly typical of the intestinal canal in advanced stages of starvation. They show the ultimate effects of starvation to consist in general atrophy of the intestine, entire removal of the epithelial coat, and extensive degeneration and destruction of the nuclei of the adenoid tissue. The degree of general atrophy may be estimated from the fact that, while in the healthy tadpoles which were examined the average length of the intestine from the liver to the commencement of the large intestine was 1·36 inch, and its average breadth 0·018, in the starved specimens the corresponding averages were only 0·57 and 0·011 inch.

The absence of epithelium appears to be invariable in cases of advanced starvation, and in them the destructive change has generally been so complete as to leave no evidences capable of explaining by what processes it has been effected. Careful observations on specimens in earlier stages of starvation, however, clearly determined that they essentially consist in a fatty transformation of the contents of the cells, followed by gradual atrophy and disappearance of the tissue which they compose. Even after very brief periods of deficient nutrition changes begin to manifest themselves in the cells. The processes of change do not, however, affect the entire extent of the epithelial surface simultaneously. They first declare themselves over an area extending from a point at some distance from the upper extremity of the small intestine to another towards its lower extremity. From this area the changes spread gradually both upwards and downwards; but, until the epithelium has entirely disappeared, differences in the degree of change can be distinctly traced in various regions, and the area primarily affected is often entirely denuded at a time when the terminal portions of the small intestine and the whole of the large one show a considerable amount of recognisable epithelium.

The first sign of change is shown by the cells acquiring a yellow tint towards their free extremities. This tint soon becomes very marked, and forms a distinct coloured band along the margin of the epithelial coat when viewed sectionally. The colouring is due to the accumulation of a thick granular matter within the cells, which is first deposited immediately around the nucleus, and gradually spreads thence throughout the cell. It consists of fat, which can be dissolved out by treatment with alcohol and ether. With regard to the precise nature of the process by means of which the altered cells are finally removed, I am unable to give any definite opinion. During the advance

of the transformation over an area of epithelium the thickness of
the layer of cells seems gradually to diminish until, just before
the complete disappearance of the coat, it is represented by a
mere narrow granular, yellow band on the surface of the reticular
tissue ; but whether the individual cells are gradually atrophied
and consumed away, or whether the phenomena are due to the
process first affecting the largest, fully developed cells, and, sub-
sequent to their destruction, invading younger less developed
structures, I was unable to determine. Whatever the precise
nature of the process may be its ultimate effect is undoubtedly
an entire destruction of the epithelium (*vide* fig. 9).

FIG. 9.—Coats of the intestine in advanced starvation, showing complete
absence of epithelium.   × 180.

The débris of the destroyed tissue appears in great part to
enter the cavity of the gut, and passes on to form an important
constituent in the evacuations which continue to be passed in
perceptible amount up to the later stages of starvation. Careful
examinations were, on several occasions, made of these excreta.
They consisted, in great part, of amorphous particles, probably
derived from dust which had entered the fluid, but they always
contained considerable quantities of soluble oily matter. The
infusoria, too, which are almost invariably present in the intes-
tinal canals of the larvæ, were generally full of oil globules and
granules in cases where their host had been starved.

The changes in the nuclear elements of the reticular tissue
are always extremely marked. As in the degeneration of the
epithelium, the process of change does not appear to occur
simultaneously over the entire extent of the intestine, and as it
does not go on to complete destruction and removal of the tissue,
differences in the degree of affection in different portions of the
intestinal surface are evident, even in the last stages of starvation.
In this coat the changes seem to occur first, and to attain their
highest development in the upper portion of the canal, while in
other parts, in proportion to their distance from this portion, they
appear later and are less complete. The earliest symptom of
change here also is a change in the colour of the affected struc-
tures. The nuclei, in place of presenting their normal soft,
colourless appearance (*vide* fig. 10), assume a yellow colour,
and become distended with a thick yellow material. This
material in some cases is distinctly granular, but in others it
appears rather as though it were a thick fluid. As the change

continues, the nucleus appears to become converted into a mere aggregation of this substance; and, judging from the presence of
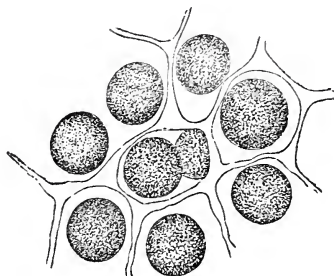
Fig. 10.—Healthy nuclei of the adenoid tissue.   × 1000.

free granules and masses of it in the meshes of the tissue, the process seems in many cases to end by the breaking up of the mass.   The material, like that in the altered epithelium, can be readily determined to be of an oily nature.

In the earlier stages of starvation such material alone is deposited in the nuclei; but in the later stages of the process it is apparently invariably associated with more or less pigmentary deposit.   The amount of pigment varies greatly in different cases, in some being comparatively scanty, in others so excessive as to form the most conspicuous feature in the tissue, and greatly to obscure the fatty deposit with which it is associated (*vide* fig. 11).
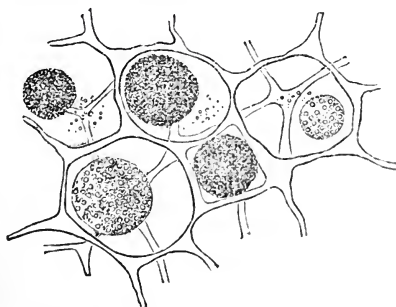
Fig. 11.—Appearances of the nuclei of the adenoid tissue in a case of advanced starvation.   × 1000.

Its distribution over the intestinal surface is similar to that of the fatty deposit, the upper portion of the small intestine being the most constant site of its extensive occurrence.   In some cases the nuclei appear to be reduced to mere masses of pig-

ment granules, and numerous free granules lie loose in the meshes of the tissue.

The extensive deposit of pigment is probably due to the great destruction of blood-corpuscles accompanying starvation. The time of its appearance coincides with that period of starvation in which distinct evidences of considerable destruction in the blood present themselves; and observation seemed even to indicate a certain amount of correspondence between the degree of blood-destruction and of pigmentation in individual cases.

Starvation also produced most marked effects on the liver. The size of the organ is greatly diminished; it assumes a dark brown colour in place of the pinkish-yellow tint normal to it; and its substance appears granular. When examined microscopically it is found to be bloodless, and its substance reduced to a mere amorphous mass of dark yellow and brown oil granules, with a sprinkling of bright red concretions scattered through it in some cases. The gall-bladder is invariably full, and very frequently is greatly distended. The contained fluid, in place of being almost colourless, as it ought to be, is green. The depth of the colour varies considerably, but in some cases is so intense as to cause the gall-bladder to appear as a deep emerald-green body, which shines prominently through the abdominal walls. The gall-bladder, in any case, usually contains some fatty concretionary matter; but in starved cases this is present in excessive quantity, and is frequently of a deep green colour.

In the case of the intestine, as in that of the tail of the larvæ, the ultimate effect of starvation consisted in a destruction of tissue associated with fatty change and subsequent disintegration of the component elements. The principal difference between the two cases was one of degree rather than of kind, the amount of destruction in the intestinal tissue being much greater than that in the tail. In both cases the destructive changes were specially manifest in active epithelial elements, that is, in structures in which the living protoplasm bore a high proportion to the amount of formed material. The occurrence of fatty change in the free sub-epidermal cells finds a parallel in that presenting itself in the nuclear elements of the adenoid tissue of the intestine, and in both cases the connective-tissue structures were little affected and remained persistent after the occurrence of extensive destruction in other parts. The phenomena in both cases, and specially those occurring in the intestinal canal, show conclusively that during the course of starvation the changes are not limited to mere atrophy, but that in some tissues extensive destruction of the component elements occurs, rendering the latter incapable of recovery on the subsequent addition of nutritive supply. So long as any living material remains within the

affected elements the possibility of recovery remains; but when they have absolutely broken up, or where they are converted into mere aggregations of dead matter, it is clear that restoration of the tissue of which they form a constituent can only be effected by the formation of new elements.

It is also clear that the amount of transformation of tissue taking place in the organism is not of necessity directly indicated by the mere chemical determination of the amount and nature of the materials leaving the body in the various excretions, for the products of transformation need not necessarily pass off, but may accumulate in large quantities, as in the case of the pigmentary and fatty deposits occurring in the intestinal tissue as recorded above.

The experiments which were conducted with larvæ of *Rana tigrina* showed that in these also the destructive effects of starvation were specially centred on the intestinal canal, and that the changes in the tissues of the mucous membrane were essentially similar to those just described.

In so far as the present experiments afford any information on the subject the changes effected by starvation in animal and vegetable tissues are fundamentally the same, and the variations presented by the phenomena attending the process in individual cases are mainly dependent on variations in the amount and character of the formed material present. Where this is small the effects of destruction and removal of the protoplasmic constituents are rendered rapidly conspicuous, but where it is present in large amount and is of a resistant nature the occurrence of change is to a great extent masked, and large portions of tissue which, in so far as active function is concerned, have in great measure ceased to serve any purpose in the organism, may, to outward appearance, remain almost unaffected.

III.—*The Conclusions drawn from a comparison of the data acquired from the Autopsies of persons who have died through want with those obtained by Experiment.*

After a detailed description of the post-mortem lesions observed by the author in the relief camps of the famine districts of Madras, which went to show that the general result of the entire series of observations was that the disease conditions under investigation were specially characterised by extreme general anæmia and destructive processes affecting the mucous membrane of the intestinal canal, the report is brought to a conclusion as follows :—We have now to consider what the phenomena observed in the post-mortem examinations of cases

of famine-diarrhœa and dysentery indicate in regard to the
essential nature and origin of the disease processes proximately
causing death.   The prominent symptoms observed during life
are those of diarrhœa and dysentery ; we must attempt to ascer-
tain whether and how far the diseases merit the titles of "famine-
diarrhœa" and "famine-dysentery."   Any forms of disease
occurring during periods of famine may, in a sense, be termed
famine-diseases ; conditions of scarcity and distress must, no
doubt, more or less favour the prevalence of all diseases.
Cholera and smallpox, for example, probably find in the famine-
stricken a most favorable field, but there is no evidence to prove
that famine alone can produce either the one or the other.   In
other words, these and other diseases are not directly due to
famine, but they may and probably do become much more pre-
valent in consequence of famine than they otherwise would have
been.   But is there evidence to show that the so-called "famine-
diarrhœa" and "famine-dysentery" possess characters, either
in their symptoms or in their pathological appearances, sufficient
to distinguish them as the results of insufficient nutrition ?
Does the evidence regarding them justify us in believing that
special forms of intestinal disease prevail during periods of
scarcity, essentially dependent on the effects of insufficient
nutritive supply ?   It appears to me that it does.   Subjects of
famine are of course exposed to the ordinary exciting causes of
diarrhœa and dysentery, and may probably be specially suscep-
tible to their influence ; but the pathological phenomena charac-
terising many cases of such disease in them appear to indicate
very distinctly that the predisposing cause is starvation, and that
the symptoms are fundamentally due to destructive changes in
the mucous membrane of the digestive canal induced by imper-
fect nutrition.

On comparing the results of the autopsies in cases of these
diseases with those obtained by experiment, it appears clear that
the changes effected by defective nutritive supply in the human
subject are closely analogous to those occurring in the amphibian
larvæ.   In both cases a fatty change and subsequent disappear-
ance of tissue elements occur ; and in both this change is
specially pronounced in the tissues of the intestinal apparatus.
The phenomena observed in the human subject show as distinctly
as those in the amphibian larvæ that the effects of famine on the
tissues are to produce an actual destruction of tissue, and not
a mere atrophic diminution of bulk, as affirmed by Bauer.   The
great diminution in the mass of the blood, which forms such a
conspicuous phenomenon in starvation, would of itself seem to
indicate that destruction of tissue elements occurs, unless it be
demonstrated that the anæmia is merely relative and solely due

to diminution of the fluid constituents of the blood; but the phenomena occurring in the intestinal canal appear to be quite decisive. Without the experimental data it might have remained a more open question whether the conditions in the human subject were not rather secondary phenomena, dependent on the fatal diarrhœa and dysentery, or due to some occult cause inducing the latter; but when taken along with the phenomena of uncomplicated starvation in the larvæ they appear unequivocally traceable to the influence of defective nutrition. The clinical experience obtained in the conduct of relief camps also affords most conclusive evidence on the question. Had all the tissues retained their integrity and capability of performing their proper functions, the excessive mortality and the general futility of the most careful dietetic treatment among cases of advanced starvation would remain utterly inexplicable.

When, however, we recognise the existence of destructive change in the tissues of the intestinal canal, these problems admit of ready solution, and all occasions to call in arbitrary explanations of the prevalence of fatal diarrhœa and dysentery among the subjects of starvation ceases. It has been usual to ascribe these symptoms to the influence of noxious materials in the blood derived from abnormal transformations of tissue, and such materials may, no doubt, be present, but the local effects produced on the intestinal tissues appear to be quite sufficient to account for the symptoms. It has also been indicated, as a curious effect of starvation, that it should produce an incapacity for the assimilation of food when supplied.[1] With the present anatomical data it would rather be a matter of wonder if it did not. With the degeneration and destruction of the epithelial and glandular elements of the mucous membrane of the digestive system, the digestion and assimilation of nutritive materials supplied in the food must necessarily be impaired or destroyed, according to the degree of morbid change. The food-elements, not being submitted to their normal transformations, become mere foreign bodies liable to undergo decomposition, and well adapted to cause irritation, especially on surfaces which have been more or less denuded of their normal protective coverings, as is the case with the intestinal mucous membrane after the destruction of its epithelium.

That symptoms of intestinal irritation should set in under such circumstances is only what might be expected, and that these symptoms should have been especially liable to occur in people shortly after admission into relief camps is readily explicable. While they were outside and actually suffering from extreme privation, the primary destruction of tissue was no doubt

[1] Carpenter; op. cit., p. 112.

advancing, but the amount of nutritive material ingested was correspondingly reduced, and was so small as to be within the control of the remaining digestive tissues. On admission into camp a larger amount of food was supplied ; the digestive and absorptive apparatus which had formerly sufficed was now relatively greatly reduced, and the surplus food-elements became mere sources of irritation. It has long been recognised that great caution is necessary in regard to food-supply after even comparatively slight starvation. Where changes in the tissues have only been slight, careful dietetic treatment, so as to avoid irritation, may suffice to tide over the danger, and ultimately effect a cure. Where, however, extensive destruction of the tissue-elements of the mucous membrane has taken place, it is clear that no dietetic treatment, however carefully carried out, can be expected to effect recovery ; dietetic treatment may save a weakened mucous membrane ; it cannot make a new one.

All the phenomena observed in the present series of investigations point to the absolute necessity of great caution in regard to dietetic experiments, dietetic systems of punishment, &c. They show that it is not safe to push such procedures in the belief that, so long as no evident active evil results present themselves, we can at any time pull up and restore things to their normal state. The fact that in so many cases the fatal diarrhœa and dysentery first manifested itself in people after their admission into the relief camps is very significant. The people in such cases were admitted into camp, showing, no doubt, symptoms of extreme general mal-nutrition, but suffering from no active symptoms of disease. The mischief had, however, been irrevocably accomplished, and it only require a change—a favorable change too—in conditions to cause it to manifest itself.

The insidious character of the mischief has a most important bearing on the practical question of the management of famines. Due to it relief camps may, to a great extent, be rendered useless by the people failing to have recourse to them until it is too late. They, too, are likely to be deluded by the idea that, where no active symptoms have appeared, no permanent damage has been done, and that they may safely delay until their distress has counter-balanced their natural inertness and dislike of disturbing their ordinary habits.

CALCUTTA ; *October*, 1878.

On the DEVELOPMENT *of the* SPERMATOZOA.   Part I. *Lumbricus.*
   By J. E. BLOOMFIELD, B.A. Oxon.   With Plate VI.

THE investigation into the structure of the testis and the de-
velopment of the spermatozoa, of which the present chapter on
Lumbricus is an instalment, was undertaken at the suggestion
and with the kind supervision of Prof. Lankester, and has been
carried out partly in the laboratory of Exeter College, Oxford,
partly in the new zootomical laboratories at University College,
London.

The true TESTES of the earth-worm were first described by
Hering ('Zeitschr. für wiss. Zool.,' vol. viii), who did not, how-
ever, figure them in position.   They occupy much the same
position in the tenth and eleventh rings of the body as do the
ovaries in the thirteenth; that is to say, they are placed in
pairs in those rings well forward, so as to be in relation to the
anterior septal wall and near the neural median line.   Each
testis is a pure white, translucent body, of irregularly quadran-
gular form, rarely more than $\frac{1}{10}$th of an inch in diameter,
much flattened, and attached by one side to the cœlomic epithe-

lium, of which it appears to be a local modification.   The
position of the testes is seen in the diagram woodcut.   Each
testis consists of a mass of spherical cells (Plate VI, fig. 1),
those at its free edges tending readily to separate and fall into
the cœlomic space or body cavity.   These readily separated cells
have, moreover, often advanced in development beyond those
which lie more deeply in the mass, and have attained the con-
dition of spermospheres or sperm-polyplasts described below.
I have not been able to determine the line of demarcation
between cœlomic epithelial cells of the ordinary kind and those
which build up the substance of the testis.   There does not
appear to be a complete investment of the testis by normal
cœlomic epithelium as there undoubtedly is of the ovary.
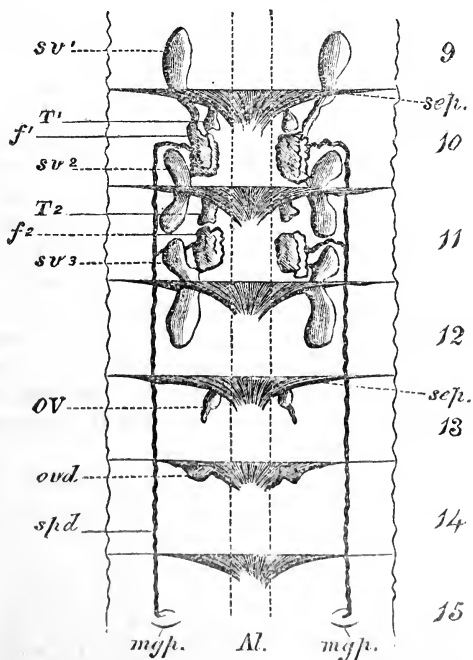
The large lobular, yellowish-white masses which are frequently
taken for the true testes of Lumbricus are, as Hering pointed
out, really *seminal reservoirs* or *vesicles,* into which the incom-
pletely developed cells thrown off by the true testes are received,
and where they undergo further development.   I am indebted
to Mr. A. G. Bourne, assistant in the Zootomical Laboratory of
University College, London, for the demonstration of the truth
of Hering's views on this subject.   Mr. Bourne showed that
the true testes are usually overlooked, owing to the fact that one
naturally selects a well-grown specimen of an earth-worm with
fully developed cingulum for dissection.   In such specimens the
periodic development of the seminal vesicles or reservoirs has
proceeded so far that they form the two well-known anterior
bilobed and posterior unilobed pairs of so-called testicular sacs,
the middle portions of which stretch across the middle line
coincidently with the septum between rings 10 and 11, and
with that between rings 11 and 12.   In this state the *true
testes* are completely hidden from view, and being at this time
completely enveloped by the enlarged seminal reservoirs cannot
be demonstrated.   Mr. Bourne, however, found in a series of
earth-worms dissected for the purpose of tracing the development
of the seminal reservoirs, when these bodies are in an incom-
plete or in the periodic undistended condition, that it is quite
easy to exhibit the four testes, as represented diagrammatically
in the woodcut here given.

The SEMINAL VESICLES or RESERVOIRS are seen in immature
specimens of Lumbricus as six small light-coloured vascular
growths on the *three* septa 9-10, 10-11, 11-12, arranged in
three pairs.   The anterior pair grow forwards so as to project
into the ninth ring, the second grow backward into the
eleventh ring, and the third pair grow backward into the
twelfth ring (see woodcut).   In the tenth and the eleventh ring
are the four ciliated rosettes or expanded mouths of the seminal

ducts, a pair in each (see woodcut). It is suggested to me by Professor Lankester that the seminal vesicles possibly originate as pocket-like outgrowths of the side-walls of these rosettes, the anterior pair carrying each an anterior and a posterior outgrowth, whilst the posterior pair have only each a posterior outgrowth. The developing sperm-cells shed by the testis are collected in the ciliated folds of the corresponding rosettes, and in all probability conveyed by their agency to the increasing seminal reservoirs. As sexual maturity approaches the three primitive pairs of seminal vesicles become larger and larger, and finally the four anterior pairs meet in one mass in such a way as to form a central body, covering in the rosettes and testes of the tenth segment, and also encroaching upon the eleventh segment; to the four corners of this central oblong body are attached four lobes corresponding to the anterior and middle pairs of the primitive seminal vesicles. A similar coalescence of the proximal portion of the posterior pair has taken place in the eleventh segment, with invasion of the area of the twelfth segment; but there are only two lobes—the backward-growing pair of vesicles which appeared on the septum between segments 11 and 12.

The minute structure of the seminal vesicles is remarkable and has not hitherto been described. From their earliest appearance they are essentially highly vascular pouches, but they do not exhibit at any time a simple cavity or lumen. The whole of the interior of the organ is traversed by vascular trabeculæ, which consist of excessively delicate connective tissue and exceedingly fine blood-vessels. The larger vessels are seen in Plate VI, fig. 4. The vascular trabeculæ form a sort of sustentaculum, in which the developing sperm-cells are packed.

To gain an idea as to the structure of this sustentaculum it is a good plan to peel off, wash, and stain a portion of the delicate tunic with which the seminal reservoir is covered. On comparing this with the sustentaculum seen in sections it is found that they agree in essential structure, and, moreover, the inner supporting portion is continuous with and formed by the indipping of the outer tunic. This outer coat of the reservoir consists of a membrane supported by fibres irregularly disposed and varying in shape. Many of them are spindle-like with a well-stained nucleus at the equator, and sometimes branching at the ends (fig 2), others are longer sinuous fibres. Among the fibres are seen a number of nuclei which, by treatment with nitrate of silver, are found to belong to the large flat cells which cover the exterior of the reservoir, and are similar to the cells elsewhere forming cœlomic lining of the perivisceral cavity.

This tunic dips down into the reservoir and forms a tra-

becular sustentaculum, dividing it into a number of small, irregular compartments, in which the true sperm cells undergo the changes requisite for the development of the spermatozoa, and accompanying these trabeculæ are seen blood-vessels and capillaries in section.

If a seminal reservoir be placed directly in alcohol, so that the red fluid is coagulated, and sections made without staining, the blood-vessels will be brought out as if they had been injected; and such sections will show that the blood is not confined to the surface, but enters into the substance of the reservoir with the trabeculæ (fig. 4); in fact, the trabeculæ are little more than networks of blood-vessels. One large vessel runs along the internal aspect of the reservoir, and from this main trunk are given off the secondary vessels which ramify over and into the organ.

Development of the Spermatozoa.

An account of the development of the spermatozoa of the lower animals has been for some time a want. While the ovary and ova have been the subject of many investigations, the testis and its contents seem not to have met with the attention they deserve; and as the earth-worm is such an easily attainable specimen, and its testicular products so easy of examination, it is a wonder that it has been so long neglected; for if a portion of the contents of a seminal reservoir are examined in salt solution, a great many of the stages of the developing spermatozoon are exhibited in one field.

The method of examination and preservation which I have found to answer best for the delicate cells is to expose them in salt solution to the vapour of osmic acid, stain with picrocarmine and mount in glycerine; and for the testes themselves and the seminal reservoirs I cut sections of the tenth, eleventh and twelfth segments, when, if all the sections are kept, the testes and also the reservoirs cannot fail to be found. Examination after treatment with osmic acid and staining reagents I have found more satisfactory than viewing them in the fresh state, as the cells are of such a delicate nature that even salt solution has a slight effect on them. A good method, when it was particularly needful to be sure that the corpuscles were perfectly unaltered, was to use the hæmolymph from the perivisceral cavity only, with the precaution that there was no confusion of the corpuscles peculiar to that liquid with the testicular cells.

As each cell of the testis itself is the source of many spermatozoa it is needful to employ terms which shall distinguish the different stages of the sperm-cells. Professor Lankester has

suggested the following terminology. The term spermatospore (parallel to oospore, denoting the ovarian egg-cell) is applied to the constituent cells of a testis, derived from the primitive germ-epithelium. These, by division of their nuclei, form spermatospheres or sperm-polyplasts. Each constituent of a sperm-polyplast is a spermatoblast, and when the process of division is over each spermatoblast becomes a spermatozoon.

The spermatoblasts, as a rule, stand out like buds from the cell which generates them, hence the name sperm-buds (spermatoblasts) applied to them. The whole of the spermatospore does not appear to be used up in the process of division to form spermatoblasts; a central or eccentrically placed portion, which may or may not be nucleated, remains passive, and serves to carry the spermatoblasts. This body, which, as will be seen below, has a central position in the earth-worm, is to be called the 'sperm blastophor' or 'blastophoral cell.' The terms thus defined find their application in a variety of different animals, and it appears probable, from my investigations on Lumbricus, Tubifex, Hirudo, Helix, Arion, Paludina, Rana, Salamandra, and Mus, that the primitive spermatospore always give rise to a passive blastophor and peripheral spermatoblasts, which latter only are directly converted into spermatozoa.

The earliest condition of the spermatospores (excluding their embryonic phases) is seen in a teazed testis. They are spherical corpuscles, averaging ·01 mm. in diameter. The nucleus is comparatively very large and possesses a well-marked nucleolus; the thin coat of enveloping protoplasm is granular, and often of a prickly appearance (Plate VI, figs. 6—10).

In a preparation of a young seminal reservoir the next stages of the formation of the spermatozoa will be seen. Many of the primitive corpuscles are seen with two nuclei, and an intermediate constriction of the enveloping protoplasm, which is often very scanty, but distinct when it is marked by granules. The thorny processes of the protoplasm may still continue (figs. 16—20). Next to this in a normal series comes the corpuscle with four nuclei (figs. 21, 22), but abnormal forms with three do occur (figs. 23). Growth of the whole cell continues, so that each of the segments in this stage may be as big or bigger than the primitive spermatospore; the nucleoli are large and distinct, often situated near the periphery of the nuclei. The amount to which the protoplasm is constricted in agreement with the nuclei varies slightly; sometimes the segments look as if they were quite separate, at other times as if they were bound and held together in an investing matrix.

The spermatosphere or polyplast, which exhibits eight segments, is the normal follower of the quadripartite spheroid (figs.

24, 25) ; but a not uncommon form is that with six, which is to be regarded as abnormal.

It is in this stage that there is first any indication that, as the spermatoblasts are being formed, a slight quantity of protoplasm is being left in the centre of the generating polyplast, which, as development proceeds, will form a cushion on which the sperm rods may rest. It is best seen in polyplasts which have been subjected to pressure, when the filament-cells or spermatoblasts will be squeezed asunder, but remain connected with the central substance by fine strands of protopalsm (fig. 26). This central mass is the 'blastophor.'

The general outline of the eight-divided cell is circular, but often oval.

The following stages up to the appearance of the complete spermatozoa may be made out in a preparation of a well-developed seminal reservoir from a large worm. The youngest stage in such a preparation is a flattened, oval, plate-like corpuscle, which, in the fresh state, appears to be composed of finely granular protoplasm, with very slight indications of the several spermatoblasts of which it consists, except a curious vacuolation, which, having a radiate direction, seems to mark the separate cells (fig. 29). These corpuscles are highly refractive, and have sharp, well-defined outlines. On treatment with acetic acid, or the mode recommended above, these corpuscles become broken up into their component spermatoblasts. Besides these, in the fresh state occur others, in which the component cells are more distinct, owing to projecting pieces of protoplasm, which give them an angular appearance.

It is not possible to determine accurately the number of spermatoblasts which form one of these corpuscles, as only one side is presented to view, but it probably varies from sixteen to sixty-four, or more. Viewed in optical section laterally, the spermatoblasts are seen to be placed on a central mass of protoplasm—the spermblastophor, which is flat (fig. 29), in accordance with the flattened shape of the whole corpuscle. These flattened platelike forms have arisen by division of the nucleus from the primitive phase of the spermatospore, which was a single nucleated cell. Though the primitive cell was spherical, yet, after its first division, when there are two nuclei and two cells, it is oblong and flattened ; the quadripartite form is flattened, and so is the one with eight divisions, or nearly so, as it is possible in these stages to count the component cells.

The most interesting point in this stage is the division of the whole spermatosphere. In the fresh state the indication of this division is very conspicuous, and as the component spermatoblasts are not distinguishable, it looks as if one large cell were under-

going primary division. The sign of this division is a cleft on both sides of the spermatosphere, not running transversely, but generally diagonally. The cleft on one side is, moreover, deeper than on the other. Sperm-polyplasts of this kind are found in varying states of division till two new polyplasts result, each consisting of eight or more spermatoblasts, which continue to go on growing (fig. 29).

I am doubtful as to whether all the sperm-polyplasts divide when they reach this state, or whether some do not continue their development without a break. I am inclined to think that this is the case, as the final form, from whence the sperm rods arise, has two shapes—either a spherical or an oval. The first would result where the corpuscle had undergone division at this stage ; the second, where there had been no multiplication of the kind.

After this division the peripheral spermatoblasts continue to multiply by fission in planes radial to the whole polyplast. The whole corpuscle increases slightly in size, and the central mass of protoplasm or blastophor begins to attain some magnitude (figs. 30—36). Henceforward it can be distinguished as a large central corpuscle.

When the spermatoblasts have reached a suitable size, the coat of protoplasm which has been enveloping the nucleus begins, in each, to collect in a small cap or knob-like mass at the distal end (fig. 37), and, from its high refractive power, constitutes a very conspicuous part of the spermatoblast, even in the fresh state. From this knob proceeds a small whip-like filament of protoplasm, at first very fine and short. It soon grows out into a lash, and constitutes the vibratile tail of the mature spermatozoon (figs. 38—42).

It does not stain readily, and requires, at all times, a good light to be seen distinctly in its early stages.

The spermatoblast at this stage may be described as pear-shaped, with the stalk of the pear turned outwards, and constituted by the whip-like filament, the body of the pear by the nucleus which joins the blastophoral corpuscle by another thin process of unstained matter.

The sperm-polyplast now is spherical or oval in shape, consisting of a number of elongated spermatoblasts, varying in amount according to the size of the sphere, supported like pins on a pincushion on the central blastophor, which has swollen up considerably, and can be seen in optical section, or even better where rough usage has rubbed some of the spermatoblasts off or flattened out the whole polyplast.

When the polyplast has reached this stage all further multiplication of the spermatoblasts ceases, and their further de-

velopment consists in a growth in length, both of the rod-like
and lash-like portion.

The pear-like shape of the spermatoblast gives place to an
elongated oval with a conspicuous knob-like head, from which
proceeds the lash-like filament. The elongated oval grows into a
short rod with a prominent cap. The short rod then grows into
a comparatively long one, till the length of a mature spermato-
zoon is attained, though the distinction of nucleus "cap" and lash
may be traced as long as the sperm rod is attached to the central
blastophor (figs. 43—48). In the mature spermatozoa the
nuclear portion is only distinguishable from the rest by its rather
greater thickness. When the sperm cells reach the stage of
mature spermatozoa they fall off from the blastophor and find
their way into the ciliated vasa deferentia. The further fate of
the blastophor it is hard to decide. It has no nucleus, but
sometimes a vacuole is visible (fig. 49). Often it is pigmented,
and it is probable that it atrophies, having served its purpose of
a support to the spermatoblasts.

The blastophor or central mass of the earth-worm's sperm-
polyplast possesses a very high interest, and will be found to be
represented, I believe, in the development of the spermatozoa of
most members of the animal kingdom. I have seen it in
Hirudo, in Helix (Pl. VI, fig. 73), and in Rana (fig. 74) and Sala-
mandra. In the two latter it is *nucleated*, a difference from the
earth-worm, which I endeavour to explain in the next paragraph.

General conclusions.—I have found no consistent account of
the development of the spermatozoa of the earthworm, though
isolated phases of the 'sperm-polyplasts' have been from
time to time noticed and figured in text-books of histology.
From observations, which I am still pursuing, upon other animals,
I am inclined to think that what we find in the earthworm is
fairly typical of a large number of other organisms, including
Mollusca and Vertebrata. The general notions on the
subject appear to have made little or no advance since
Kölliker's paper, published in 1856 ('Zeitsch. fur wiss.
Zool.,' Bd. vii), which is, as might be expected from its date,
wanting in accuracy. Kölliker gives very unsatisfactory
drawings of the development of the spermatozoa of the bul-
lock, the pigeon, the frog, and the carp. His 'mother-cell'
corresponds to my 'polyplast,' but he complicates the whole
history by erroneous notions as to endogenous cell formation
and the conversion of nuclei into spirally rolled spermatozoa.
The result of my observations is that, to begin with, the *nucleus
of the primitive cell or spermatospore* in the young testis *is of
unusually large size*, and that the secondary nuclei to which it
gives rise stand out around the central mass or blastophor of the

generating spheroid with very little protoplasm clothing them. *The nucleus undoubtedly becomes the rod-like head of the earth-worm's spermatozoon,* and the filament is as undeniably formed from non-nuclear protoplasm.   The sperm-blastophor of the earth-worm is not nucleated, and it atrophies and disappears after it has shed its crop of spermatozoa.   This must be brought into relation with the fact that the development of the spermatoblasts proceeds *not* in the testis itself, but after the spermatospores have been shed from the testis and taken into the seminal reservoirs.

On the other hand, we find in the frog and salamander that what corresponds to the blastophor is a *nucleated body* (see fig. 74). The blastophoral nucleus was indeed seen and figured by Kölliker in fig. 5 v of his memoir.   In the frog and salamander the nucleated portion of the blastophor (which in these animals is *not* central but lateral) *remains adherent to the wall of the seminal tube or crypt,* and only a portion of the corpuscle breaks off, carrying with it the elongated nuclei, which become heads of spermatozoa. Thus, in the frog and salamander, a portion of the sperm-poly-plast, the nucleated blastophor, remains every year in the period succeeding the breeding season, and is very probably ready to resume its activity and produce a new crop of spermatozoa after one crop has been cleared away.   These nucleated blastophors are seen forming the lining of the testicular tubes in the frog. On the other hand, the primitive testis cells or spermatospores of the earth-worm *pass away from the testis into another organ* (the reservoir) in order to undergo their development; the whole mass of the sperm-cell is detached from its site before the blastophor and spermatoblasts have been differentiated.   Hence the *central* position of the blastophor and its temporary, evanescent character.

There is another kind of corpuscle which occurs in preparations of a well-developed seminal vesicle (reservoir) of an earth-worm, and for a long time it was a puzzle to connect it with any part of the previously described history of a sperm-cell, but the final conclusion to which I have arrived is that it has no connection with it at all.   What the function of these cells in the reservoirs is I do not know; it is possible that they form a kind of packing material during the growth and development of the spermatozoa, and may be connected with the nutrition of the developing cells and the periodic atrophy of the vascular framework of the reservoir.

In size these cells vary, but attain in the larger specimens a diameter of $\frac{1}{800}$th inch, and may be called, even when pale in tint " the brown corpuscles."   They have very generally a brown

colour due to granules. In shape they would be flat plates of an oblong outline, were they not generally bent on themselves and curved in various places. Often they are very much elongated, the longitudinal dimensions exceeding the transverse four or five times.

An interesting fact in regard to them is that they are composed of two distinct substances, one a hyaline transparent groundwork of a refractive index, very little differing from that of water, in which is held an irregular network of a denser substance with large granules, many of which, as shown by Klein for intranuclear granules, are the ends of rods of the network in optical section; but this is not the case with all. In one part of the cell, varying in position, but in the corpuscles of an elongated shape, generally situated at one of the ends, is a nucleus: sometimes two. Of these nuclei there is little indication in the fresh state except that at a certain spot the protoplasm is slightly more dense and opaque in appearance (figs. 51—59). Under treatment with osmic acid and picrocarmine the rods of the network split up into granules, often not quite completely, so that a moniliform outline is the result (figs. 60—65).

Salt solution causes a slight shrinking of the network.

The effect of water is instructive. When it has been added to a preparation in salt solution, the first effect is that the network loses its clearness, but continues to be visible for some little time. Then the corpuscle begins to swell until it becomes a hyaline sphere, containing granules which dance in rapid Brownian motion, and the nucleus is exposed to view. Dilute acetic acid has the same effect (fig. 66).

Osmic acid, when applied in solution and picrocarmine, swell up the corpuscle in a similar way owing to their water, but when glycerine is applied (as is usual for preserving the preparation) it is reduced again in size.

The vapour of osmic acid causes the network to disappear, leaving granules in its place.

The proportionate amount of this network varies a good deal. In some corpuscles the meshes are large and distinct and the rods of large dimension, but in others the meshes are small and irregular, and often the rods very fine.

At all times the arrangement of the network is hard to determine accurately, and it is only possible to give an idea of it in a drawing by diagrammatic treatment.

Of the function of these corpuscles I can only, as I have said, make a guess. The stages in the development of the seminal reservoirs in which they are found are the following: in large well-developed reservoirs they are large and numerous, but in reservoirs taken from those large dark worms, which seem

to be in a state of decline after the performance of the generative function they are *particularly numerous in proportion to the other corpuscles.* The actual number of them in this case may not be increased, but owing to the absence of the other cells which have developed into spermatozoa it seems that there are more of them. This fact seemed to indicate that they have a connection with the blastophoral corpuscle, but a fatal objection to this view is that the latter possesses no nucleus, and would have to go through a long series of changes before it would resemble one of these ' brown corpuscles,' of which intermediate phases there are no indications whatever. I have repeatedly searched for such phases and have never found them. In the very youngest stages of the reservoirs there is no sign of them, but in preparations where the sperm-cell segmentation has reached the binary stage it is seen that they are represented in a young condition, developing together with the true spermatoblasts and the fusiform cells of the reticulum. They are in this stage much smaller than the mature ' brown corpuscles,' and the network is so dense that it is impossible to make out its arrangement in detail. I have represented them at this stage in figs. 67—71.

On the SPINAL NERVES *of* AMPHIOXUS.   By F. M. BALFOUR,
    M.A., F.R.S.   Fellow of Trinity College, Cambridge.

IN an interesting memoir devoted to the elucidation of a
series of points in the anatomy and development of the Verte-
brata, Schneider[1] has described what he believes to be motor
nerves in Amphioxus, which spring from the anterior side of the
spinal cord.   According to Schneider these nerves have been
overlooked by all previous observers except Stieda.

I[2] myself attempted to show some time ago that anterior
roots were absent in Amphioxus ; and in some speculations on the
cranial nerves, I employed this peculiarity of the nervous system
of Amphioxus to support a view that Vertebrata were primitively
provided only with nerves of mixed function springing from the
posterior side of the spinal cord.   Under these circumstances,
Schneider's statement naturally attracted my attention, and I
have made some efforts to satisfy myself as to its accuracy.   The
nerves, as he describes them, are very peculiar.   They arise
from a number of distinct roots in the hinder third of each
segment.   They form a flat bundle, of which part passes up-
wards and part downwards.   When they meet the muscles they
bend backwards, and fuse with the free borders of the muscle-
plates.   The fibres, which at first sight appear to form the nerve,
are, however, transversely striated, and are regarded by Schneider
as muscles ; and he holds that each muscle-plate sends a process
to the edge of the spinal cord, which there receives its innerva-
tion.   A considerable body of evidence is requisite to justify a
belief in the existence of such very extraordinary and un-
paralleled motor nerves ; and for my part I cannot say that
Schneider's observations are convincing to me.   I have attempted
to repeat his observations, employing the methods he describes.

In the first place, he states that by isolating the spinal cord
by boiling in acetic acid, the anterior roots may be brought into
view as numerous conical processes of the spinal cord in each
segment.   I find by treating the spinal cord in this way, that
processes more or less similar, but more irregular than those
which he figures, are occasionally present ; but I cannot per-
suade myself that they are anything but parts of the sheath
of the spinal cord which is not completely dissolved by treatment
with acetic acid.   By treatment with nitric acid *no such processes*

[1] 'Beitrage z. Anat. u. Entwick. d. Wirbelthiere,' Berlin, 1879.
[2] "On the Spinal Nerves of Amphioxus," 'Journ. of Anat. and Phys.,'
vol. x, 1876.

are to be seen, though the whole length and very finest branches of the posterior nerves are preserved.

By treating with nitric acid and clarifying by oil of cloves, and subsequently removing one half of the body so as to expose the spinal cord *in sitû,* the origin and distribution of the posterior nerves is very clearly exhibited. But I have failed to detect any trace of the anterior nerve-roots. Horizontal section, which ought also to bring them clearly into view, failed to show me anything which I could interpret as such. I agree with Schneider that a process of each muscle-plate is prolonged up to the anterior border of the spinal cord, but I can find no trace of a connection between it and the cord.

Schneider has represented a transverse section in which the anterior nerves are figured. I am very familiar with an appearance in section such as that represented in his figure, but I satisfied myself when I previously studied the nerves in Amphioxus, that the body supposed to be a nerve by Schneider was nothing else than part of the intermuscular septum, and after re-examining my sections I see no reason to alter my view.

A very satisfactory proof that the ventral nerves do not exist would be found, if it could be established that the dorsal nerves contained both motor and sensory fibres. So far I have not succeeded in proving this; I have not, however, had fresh specimens to assist me in the investigation. Langerhans,[1] whose careful observations appear to me to have been undervalued by Schneider, figures a branch distributed to the muscles, which passes off from the dorsal roots. Till the inaccuracy of this observation is demonstrated, the balance of evidence appears to me to be opposed to Schneider's view.

[1] 'Archiv f. Mikros. Anatomie,' vol. xii.

The Bacillus *of* Leprosy.   By G. Armaner Hansen.
With Plate VIII.

It was not my intention to make any of my investigations on this subject public at present, but as not only Dr. Edlund, to whom in the preceding year I showed preparations, and mentioned that I considered leprosy a parasitic disease, in his little work on ' Leprosy ' speaks of its precise origin as something that he has discovered in the form of " micrococci," but also Dr. Neisser, of Breslau, who passed some portion of this summer in Bergen has just published the result of his investigations of those preparations that he made while here, and as these results also point out that in general, the preparations are filled with " bacilli " which he supposes to be peculiar to leprosy, and as its " contagium "—I feel myself called upon to announce what I have attained to, up to the present time, in my researches after the same " contagium," and, this, partly to assert my priority with reference to this discovery, and partly in order to advance those details in research which I omitted to announce on account of the still uncertain result in my report to the Medical Society in Christiania, 1874, concerning my investigations into the etiology of leprosy.   In this report I have briefly stated that I often, indeed generally, found, when seeking for them in the leprous tubercles, small rod-shaped bodies in the cells of the swelling,[1] whilst, on the contrary, I never found such " bodies " in blood newly taken from leprous patients, in which, however, Dr. Edlund declares he has seen the " micrococci " described by him.   This observation of Dr. Edlund I must, however, after having examined several times, quite lately, blood taken from a leper, consider as unreliable.   I found very often, on the contrary, after preserving the blood-preparations in a damp room, that in the course of a few days there appeared articulated threads, which, I believe, must be considered as a fungous growth, and which never appeared in blood-preparations taken from either healthy or syphilitic people.   After having employed myself for a lengthened period in these investigations of the blood, I proceeded to those of the tubercle, and shall communicate, as follows, a few of the memoranda I made during that time.

Case No. 755. — Johannes Gül, vigorous nodules; February 28th, 1873.   A nodule taken from each side of the nose with scissors, and laid in a carefully cleaned watch-

[1] *Vide* ' British and Foreign Medico-Chirurgical Review,' April, 1875.

glass. Cut through them; no softening; scraped the surface with the edge of the knife, and placed the parts removed on an object-glass, and without any addition of fluid covered over with a glass cover. There were seen almost only round-cells, very few with granules of fat, some finely granulated, others containing rod-shaped bodies, which are sometimes bordered by parallel lines, and sometimes pointed at both ends. In this latter instance they are about twice as thick in the middle as the others. Such bodies are to be found where fluid-spaces are formed by the pressure of the glass cover surrounded by the dense mass of cells. In these spaces the bodies move after the manner of " bacteria." Other preparations prepared in the same way were examined in a drop of distilled water with Hartnack No. 9, but with no result. The round cells are the regressive elements of brown colour, which I have both described and drawn in my previous contribution to " Leprosy's Characteristics," ' Nord. Med. Art.,' vol. i, No. 13; these drawings are reproduced in " Leprous Diseases of the Eye," by O. B. Bull and G. A. Hansen, Christiania, 1873. In the next place I scraped the surface of the tubercle as above, and put the scrapings into a drop of water. In such preparations an unusually large number of the small bodies show themselves, which have besides more or less lively movements. The cells mostly swell up considerably in the water, and in the swollen cells the rod-shaped bodies are much more easily found; many cells show themselves completely loaded with them; at the first glance it seems as if the cells were filled with coarse granules, but on closer examination, on the contrary, these apparent granules are found to be small, oblong, rod-shaped bodies. Several preparations of each kind are then placed on the bottom of a glass basin which is covered by a larger one; the bottom is covered with damp sand, and over all is placed a glass plate (two preparations with water, three without).

March 1st.—No examination of the preparations.

2nd.—Preparations appeared as on February 29th.

3rd.—In one of these preparations with water a mass of articulated threads was to be found in one place exactly corresponding to those which are found in the blood of certain lepers after cultivation.

4th.—In such preparations the articulated threads are to be found; in one of them without water they were found at the edge of the preparation, as well as in the spaces filled with serum amongst the densely packed cells; these were not long threads, only two or three joints fixed to each other,

and swimming partly free in the fluid. In one part of one of the preparations there was a countless number of oscillating rods, and one "penicillium." Two of the preparations without water did not fill the whole space under the glass cover; in the water which had fallen in drops in the vacant space no trace was to be found of either bacteria or anything else.

7th.—Kristian Lotuft. Vigorous tubercles which have steadily increased in the last year. One tubercle was removed and divided; one half was immediately placed in 1 per cent osmic acid, and preparations were made of the other, and the blood oozing out from the cut surface was collected in a small glass vessel.

Preparation, No. 1.—Serum containing blood-corpuscles; from this preparation a particle of the tubercle was teased out with needles, and examined under the microscope; numerous oscillating rods and rod-like bodies were to be observed in some of the cells.

Preparation No. 2.—Serum containing blood-corpuscles; here and there a white body; extremely few oscillating granulated masses.

Preparation No. 3.—Serum with numerous blood-corpuscles, and admixed "epithelium" from the edge of the cutting; numerous bacteria. All these were kept in a moist granulated chamber.

Preparation No. 4.—In a drop of distilled water pieces of the picked-out tubercle; numerous bacteria; immovable bacteria in the cells. The preparation was enclosed with oil along the edges of the glass cover.

Preparation No. 5.—Serum containing blood-corpuscles, enclosed with oil as No. 4.

Preparation No. 6.—Serum containing blood-corpuscles, with pieces of the picked-out tubercle also enclosed with oil, and containing numerous bacteria.

8th.—Nothing to remark.

9th.—No. 1. Leptothrix; articulated threads abundant.

No. 2. Nothing.

No. 3. Nothing.

No. 4. An occasional articulated thread here and there.

No. 5. Nothing.

No. 6. Like No. 4.

10th.—No. 1. Still more chains in large groups.

Nos. 2 and 3. Nothing.

Nos. 4, 5, and 6. Like those of yesterday.

18th.—Case No. 705. A tubercle taken from the under lip; softening commencing.

Preparation No. 1.—Procured after incision by pressure on the tubercle which is tolerably juicy, contains a considerable number of large brown elements and numerous bacteria ; these could also be indistinctly seen within the cells which are not of a very deep brown, and I think now and then that I could distinctly see in these a long stripe in the apparently granulated masses.

No. 2. Like No. 1

No. 3. Like No. 1 (with water). The cells for the most part were swollen, and in these rod-shaped bodies in large numbers were distinctly to be seen. The large brown elements were not much affected by water, occasionally a little swollen, and thus it can be tolerably clearly seen that in general a large portion of the apparent granulations are oblong and rod-shaped.

All these preparations were kept in a moist chamber. The addition of acetic acid does not help at all. The preparations become more opaque by coagulation.

With acetate of potash all the oscillating rods are killed ; they become instantly more highly refractive after it has penetrated, shrivel, and lie exactly like corpses all over the preparation ; the brown ones shrivel considerably, and become exceedingly refractive, shining like wax. Their whole contents become as if kneaded together ; no rods to be distinctly seen unless action be just about to commence. The rod-like bodies in the cells are to be seen best by teasing out a piece of the tubercle in 1 per cent. " osmic acid."

20th.—In many places in all the three preparations articulated threads of greater or less extent are to be found ; in some places a single thread wound a few times round itself, in others such a confusion of threads that to follow them was impossible.

Of the tubercle which, after obtaining the above-mentioned preparations on March 18th, was laid in 1 per cent. osmic acid, a preparation was taken to-day, which showed rod-shaped bodies in most of the cells. I took a drop of the reduced acid in which the tubercle was lying ; no oscillating bodies were to be found ; a small particle of the tubercle was placed into this drop and teased out, and on investigation many oscillating rods, of a length of 0·0015—0·006 mm., were to be found. On repeated knocking on the cover glass until almost the whole of the above particle had fallen to pieces, it was found swarming with oscillating rods, and in the broken edge of a large cell which had been fractured by the knocking a few rods were seen protruding into the fluid.

21st.—Numbers of articulated threads in all the preparations like those of yesterday.

April 1st.—Olive Bjorhaug. Spots six weeks old ; strong retrogression. A piece cut from a spot on the forearm, divided into two pieces, one teased out in fresh water, one in salt water ; the sudoriparous glands appear large and easily teased out; round cells amongst the windings of the canals, also here and there, between the bundles of connective tissue, many cells interwoven with strongly granulated " protoplasma." In both preparations, principally along the edges of the fragments of the teased-out sweat-ducts, a quantity of pale, round, angular and oblong molecules, with oscillating movements, were found. It cannot be positively decided whether some of the more lengthy ones were really bacteria ; highly refractive, round, and oblong small bodies could be found in some few of the sweat-ducts ; kept in a moist chamber. Besides this a preparation in osmic acid was made of the blood oozing out from the cut surface of the spot.

3rd.—In the blood preparation, which is almost dried up, numerous articulated threads were found. In the other two, along the edge of them, numerous large bacteria with lively movements, but no articulated threads.

4th.—The blood preparation quite dried up. The two others like those of yesterday, although the bacteria were not in such lively movement.

7th.—The preparation with water dried up ; the preparation with salt water, connective tissue and cells unchanged. No bacteria to be discovered; on the contrary, numerous highly refractive, small articulated threads were found, with from two to five articulations, immovable.

17th.—Christian Loluft. Obtained a preparation from a tubercle on the cheek by puncture, principally blood, with some greater or smaller tubercle cells ; unable to discover any free bacteria.

18th.—The red blood-corpuscles a little shrivelled ; the cells, however, about normal, some of them a little discoloured.

20th.—The cells extremely pale and somewhat shrivelled, and occasionally hydropic ; no bacteria to be seen in them ; the large pale brown cell-contents unchanged ; nothing more to be discovered than on the first day ; granules and doubtful rods.

22nd.—Chains in many places, but the preparation not to be relied on, on account of water having penetrated in one place.

Anne Sahingetar, died March 10th ; post-mortem examination 11th March, 1873. Some rather shrivelled tubercles from the face were examined ; large, dry brown bodies were found in parts, formidably large, and easy to be seen with the naked eye. In the microscopical preparations oscillating rods were to be found everywhere, and after the addition of potash-lye I was fortunate enough to see in several of the large brown elements a sort of striation amongst the apparent granulations ; the brown ones adhered strongly to the glass, and when the glass cover was lifted up and removed from one place to another, many pieces were found remaining on it in many places ; these pieces showed themselves to be composed of small rod-shaped bodies, which crossed each other in every direction.

March 17th.—No articulated threads have formed themselves in any of those preparations which were kept in a damp room.

21st.—Christian Loluft. Eruption over the whole body rather severe ; two newly-formed tubercles were punctured with a needle ; a drop of something resembling "pus" was squeezed out at the same time ; the drop was viscid, tough, and did not float out by pressure on the glass cover ; only a few cells at the edges became free by the addition of distilled water, also when the glass cover was repeatedly lifted up and set down on the one side. It was also evident from microscopical examination that the preparation contained a few blood-corpuscles. After the addition of water and the glass cover still remaining removed, nothing remarkable is to be found in the preparation ; if, however, on the contrary, the glass be moved from side to side so that a portion of the preparation is floated, a not inconsiderable number of oscillating rods of different sizes make their appearance in the fluid. Two similar preparations were made, one with and one without water (in a moist chamber) ; besides these a preparation of blood was made, in which, on examination with Hartnack No. 11, no bacteria could be discovered ; finally, two other preparations of the contents of the tubercle without water.

23rd.—No articulated threads to be found in any of the preparations ; those without the addition of water still appear as quite fresh.

24th.—The same as above.

25th.—Also the same.

28th.—Still no fungus. The preparation without water kept remarkably well. The cells had a perfectly fresh appearance. The preparation containing serum was also

perfectly dried up along the edge, so no water could penetrate.

29th, 30th, 31st.—On account of other engagements the preparations were not examined.

April 1st.—Nothing is seen in the blood preparation, which is mostly dried up; fungus in both the others. The preparation without water, especially deserves attention; here right in the middle of it, with so especially well-preserved cells that they appear perfectly fresh, were found in four places large masses of fungus, of finely granulated appearance, from the edge of which there shoot out fine articulated threads, whose several joints measured 0·0006—0·0007 mm.

4th.—This preparation exactly like that of the 1st. The masses of fungus have not become larger; a rather large brown one, which is broken on one side, remained unchanged in these three days. Its place was carefully noted in order to watch if any articulated threads should grow out of it. Along the edge of the glass cover, upon the dried up edge of the preparation, a brown fungus has shot out a rich network of threads with fructification, by division of the points.

7th.—The fungus mass along the edge of and in the preparation is unchanged, the edges set free, and alcohol and ether added, which only, by repeatedly lifting the glass cover, penetrate the preparation; by this the fungus mass was divided into lumps; little by little, by the action of the ether, they took the same appearance as the brown ones, viz. the granules and rods were kneaded together into a shining wax-like mass.

Hospital, Johannes Gül.—Two tubercles from the nose, taken out with entire epithelium.

Preparation No. 1.—Scraped with a knife the mass of cells in the blood-serum which lies on the surface; obtained several doubtful bacteria with slow movements.

Preparation No. 2.—With a glass tube, freshly blown, from an incision in the tubercle teased and sucked out small pieces of the cell mass; considerable number of blood corpuscles, also bacteria.

Preparation No. 3.—In like manner; fewer blood-corpuscles.

Preparation No. 4.—Ditto, with as much water; much more numerous bacteria, and here and there in the swollen cells rod-shaped bodies; many brown ones in a lump, none floating.

Preparation No. 5.—Preparation of blood collected in a

watch-glass; serum containing blood-corpuscles, here and there a white blood-corpuscle; discovered a few bacteria. On teazing a preparation in osmic acid I find many cells, which enclose longish rods.

10th, Nos. 1, 2, and 3.—Present no alterations, except that where the longish rods can be seen they are motionless.

Preparation No. 4.—In many of the cells that have been swollen by water are found more or less numerous granules of various sizes, showing molecular movement; in some of the cells between the oscillating granules a rod with slower movements can be observed, which appears independent of the movements of the granules; in other cells no granules are to be found; on the contrary, many rods and one or two of these with slow serpentine movement.

Preparation No 5.—No alteration, except that the blood-corpuscles are more contracted.

11th.—The preparations are unchanged.

12th.—The same as 11th, only that the cells have no longer anywhere the same uninjured appearance as before; in many the small rods are not to be seen.

14th.—In No. 1—3 the cells are still well preserved. The nuclei stand more boldly out than before, more homogeneous. In the dense crowd of cells is found, in many places, what appears to be a film over the preparation; this film seems to consist of a finely dotted mass, and also of small rods, which cross one another in all directions. In No. 2 is found in the serum two small chains of monads. In one place in No. 3 a well-defined granulated mass, which at the edge shows itself to be a number of chains of monads. In No. 4 only a few of the cells are preserved in one side of the preparation; an immense number of bacteria right in the middle of the preparation; in several places a mass of immovable rods crowded together. It is difficult here to ascertain the nature of these, for in many places are found small lumps of stearine crystals, but the former are found to be much more highly refractive and of a more irregular arrangement than the stearine crystals.

18th, Nos. 1—3.—The cells have disappeared, best preserved in No. 3; the finely granulated fungus mass not particularly increased, coloured a strong brown black with osmic acid, like the brown bodies. No more chains of monads in Nos. 1, 2.

20th, Nos. 1, 2.—The cells always more fallen away and kneaded together in a mass, in which the single cells

could not be distinguished. No 2 best preserved, still no chains. In No. 4 are masses of bacteria; at the edge a large mass of zoogloea, has made its appearance, amongst which are found a few penicillium threads, so at all events a part of the bacteria may be supposed to have come in from the outside, but this bacteria-containing zone has not encroached farther into the preparation than before.

21st.—In Nos. 1 and 2 in many places, in the middle of the preparation, masses of zoogloea.

March 24th, 1873.—Ratel Espeland. Eruption. Punctured an umbilicated tubercle that had become tender by the eruption; blood and the whitish contents collected. Two preparations were made. In both gigantically large brown elements to be found; no bacteria. Water was added to the one preparation. As long as this flows quietly in, and the glass cover be not moved, no bacteria appear, whilst, however, a large portion of the cells swell up; by moving the glass cover up and down, and pushing it a little to one side, a large portion of the cells break, and now bacteria appear in the fluid.

25th.—Preparation unchanged.

27th.—In the preparation to which water has been added numerous articulated threads are especially remarkable, issuing from a brown body; the connection confirmed by the movement of the glass cover. By these means it was ascertained that the whole mass in the lower element, together with the apparent articulated threads, remained connected in all positions of the former (fig. 5).

April 18th.—Ratel Espeland. From a puncture of a tubercle on the forehead; pressed out contents of it; this, so firm that it must be teased out with needles. One preparation was made in salt solution, and one in distilled water; in both an immense quantity of bacteria; the cells large. In the preparation with distilled water bacteria are to be seen in almost all the swollen-up cells, some with rather lively movements. The situation of the bacteria within the cells ascertained in the most evident manner, by getting the cells to roll in the fluid; no granules for the most part can be found in the cells, and where such is the case, they oscillate with far greater speed than the slow-moving bacteria.

20th.—Many bacteria can be found in the water preparation, which are those with 3—4—8 articulations. In the salt-water preparation the cells are not a little shrivelled; by the addition of water a large portion of them are very quickly set free; a crowd of granules stream out in

greater or smaller clumps, amongst these a number of small rods.

22.—In both the preparations abundant masses of zoogloea.

April 10th.—From Iver Sorlide were taken three samples of blood from the cheek, in freshly blown capillary tubes, which were secured at one end and hung up on a string in the room.

20th.—The contents were examined, coagulated in all three tubes, and were difficult to get out. The blood-corpuscles had a healthy appearance; the red ones rather spherical, the white, for the most part, kneaded together in clumps. In one place in one tube in the coagulation was found a mass of zoogloea; in the other two nothing could be discovered.

From the various notes of my investigations in 1873 every one will be easily able to see that I had good reason for supposing that bacteria appear in leprous products, but also that I, supported alone by these investigations, could not propound a theory on this subject, and still more decide whether these bacteria really were the virus which, introduced into the system, produced the disease. In order, if possible, to arrive at a decision in this matter, I tried to inoculate rabbits with leprosy by introducing portions of the leprous growths, especially of the tubercles, under the skin of the animals. I was not lucky in any of these attempts, which, however, as a matter of course, is a proof against the supposition that the above-named bacteria are the real virus. I have not repeated this attempt later, and only now and then, by examining the tubercles, have become convinced that my observations with regard to the occurrence of the oscillating rod-shaped bodies were correct, until I, on reading this spring Dr. Koch's work, 'Untersuchungen uber die Ætiologi der Wund-infections krankheiten,' and by being enabled to see some of Dr. Koch's preparations of *Anthrax bacilli*, learned to know this author's excellent system for the demonstration of bacteria. It occurred to me this summer to try ether in the investigations, in order, if possible, to prove the bacterium-nature of the rod-formed bodies, and their presence every-where where leprous productions are to be found. But I have hitherto endeavoured in vain to obtain good and convincing preparations, except in one case, when I was able to obtain perfectly convincing results. Guided by the above-recorded observations, viz. that the small rods become more distinct by the treatment of the tubercles with osmic acid, I placed an extirpated tubercle in this acid, and have

obtained from it, by colouring the sections with methyl violet, a few preparations in which the brown cells, easily visible, also without colouring, appeared very sharply distinguished from the surroundings by their violet colour; upon closer inspection they show a finely granulated and partly striated appearance, as if they consisted of small rods. As a reason for my small success in this work I must (after a letter from Dr. Koch, with whom I communicated respecting it) presume that the reason is either that the methyl violet I use is not as it ought to be, or that I have allowed the colouring matter to act too short a time. These mistakes, Dr. Koch informs me, were from the first committed also by Dr. Neisser. Dr. Neisser's later and more successful results probably arise from his having been so fortunate as to have Dr. Koch's valuable guidance. I have, by this preparation, obtained conformation of my earlier supposition that the large brown bodies after all are nothing else than either masses of zoogloea or collections of bacilli which are enclosed in cells. By looking at fig. 4, which represent tumour-cells treated with osmic acid, drawn from preparations made in 1873, one is easily able to form an idea how these same cells, by a constantly increasing number of small rods, at last become quite overloaded, and thus obtain the appearane of being filled with fine granules, since the single rods cannot then be distinguished. I have already in my first communication to "Leprosy's Characteristics" ('North. Med. Archiv,' vol. i, No. 13) stated that I was inclined to regard those brown elements as peculiar to leprous growth, both on account of their most striking appearance, and because they were always to be met with in all the parts affected with leprosy. Should the above-mentioned supposition concerning the true nature of the brown elements prove in time to be correct, their peculiarities will, at the same time, be demonstrated, and it will be important to ascertain the conditions of existence of these bacilli, in order, finally, with full reliability to remove all doubt as to the real cause of leprosy; and this, shall as before, be the goal of my work.

Since writing the above I have also been so fortunate as to obtain bacilli finely coloured in a section of a tubercle hardened in absolute alcohol, and, acting upon Dr. Koch's advice, stained with a stronger staining fluid. Baccilli are found in all parts of the section, either singly, or more frequently in groups, fully corresponding to those occurring in the cells. I furnish a drawing of two groups taken with Zeiss's immersion system $\frac{1}{12}$th, and eye-piece No. 4.

NOTES AND MEMORANDA.

Development of Planorbis.—Dr. Carl Rabl, of Vienna, has published in Gegenbaur's 'Morphol. Jahrbuch,' vol. v, a very carefully illustrated account of the development of the fresh-water snail Planorbis. In essentials the history does not differ from that of Limnæus, on which subject Dr. Rabl some time since published a less complete and less accurate memoir ('Jenaische Zeitschr.,' 1875). In the present memoir Dr. Rabl abandons his erroneous interpretation of the shell-gland and recognises the correctness as well as the priority of my descriptions of that organ in the Mollusca generally and of the velum of Pulmonata. Dr. Rabl has, however, failed to trace the derivation of the hinder part of the embryonic intestine from a group of cells adhering to the margin of the blastopore (constituting a " pedicle of invagination " as in the Lamellibranchs Pisidium and Anodon); on the contrary, he considers this portion of the enteric tract to be a cæcal outgrowth of the invaginated endodermal sac, which outgrowth pushes forward so as to touch the body-wall. The endodermal sac (archenteron) is only in contact with the blastoporal margin (according to Dr. Rabl) at the anterior or oral end of that orifice. Satisfactory evidence of this is not adduced. Drawings of a very beautiful series of sections are given in two plates as well as numerous transparent views of young embryos. The drawings of sections are perfectly accurate, as I am able to state from the examination of a number kindly sent to me by Dr. Rabl. The sections are masterpieces of embryological manipulative skill, but they do not tend to substantiate the view with regard to the origin of the hinder part of the embryonic intestine which Dr. Rabl maintains. They are equally favorable to the view to which I was led by observations recorded in this Journal in 1874. Dr. Rabl takes so strongly adverse a position in reference to the connection of the hind-intestine with the blastopore, that he emphatically denies the accuracy of my observations on the connection of the blastopore and anus in another Gasteropod, viz. Paludina.

He is fully aware of the fact that Professor Bütschli examined the development of Paludina ('Zeitsch. wiss. Zool.,' vol. xxix) for the express purpose of testing the accuracy of my statement, and came to the conclusion that the matter was as stated by me.   Dr. Rabl is scarcely well advised to impugn the correctness of an observation, thus supported, *without offering illustrative drawings*, showing what it is that he has seen in the history of Paludina.

The formation of the primitive nephridia of the embryo Planorbis, by perforated cells, similar to those forming the nephridia in adult Lumbricus and Hirudo, is admirably shown by Rabl's sections and drawings.—E. RAY LANKESTER.

Origin of Sperm and Ova from the Cell-layers of Cœlentera.—The researches of the brothers Hertwig (' Organismus der Medusen,' Jena, 1878) upon the histology of a number of Medusæ belonging to the group known as Hydromedusæ led them to the conclusion that, uniformly in Medusæ of that group, the genital products are derived from the Ectoderm. This conclusion, corresponding with that of Kleinenberg relative to Hydra, led the Hertwigs to consider it probable that the observations of Ed. van Beneden, Ciamician, and others, which attributed the ova, or, in some cases, the sperm to Endoderm in the sporosacs of certain hydroid polyps, were erroneous, and that the derivation of the ova and sperm from Ectoderm might be considered as general for all the Hydrozoa.

Claus, however, found the spermatozoa of the medusa Chrysaora developing directly from cells of the Endoderm over a large surface of the gastric pouches (Polypen und Quallen der Adria, ' Wiener Denkschr.,' 1878), and now it appears, from further researches of the Hertwigs, upon Lucernaria, Pelagia, and Charybdœa (' Jen. Zeitschr.,' vol. xiii, 1879), that in the Medusæ of the Lucernarian type (Acraspedæ of Gegenbaur, or better called Scyphomedusæ, as opposed to Hydromedusæ), the ova as well as the spermatozoa uniformly take their origin from cells of the Endoderm.   This being the case it seems quite possible that, after all, both the observations of Ed. van Beneden and those of Ciamician may be accurate, and that in the Hydrozoa there is no absolute uniformity as to the origin of the generative products.   This, however, is not the view taken by the Hertwigs.   They have extended their observations to certain—a very limited selection—of the Anthozoa, and find that in them (various Actiniæ, Cerianthus, and Alcyonium)

the ova and the spermatozoa both take their origin from cells of the Entoderm. Accordingly, the Hertwigs propose to generalise their observation—a proceeding which appears to us to be as yet without justification. They still maintain—in spite of the observations of Ed. van Beneden and others—that *all* the Hydromedusæ develop their genital products from Ectoderm. This really rests only on the observation of a certain number of medusa forms and of Hydra, and is opposed to equally careful observations on Hydractinia, &c. Further, they expand their observations on Scyphomedusæ (Acraspeda) and Anthozoa to the dimensions of a statement that *all* members of both these groups uniformly develop their genitalia from Endoderm. Very possibly both these large statements are true, but such is by no means demonstrated at present. However, the Hertwigs proceed further, and on this basis propose to remodel the groups of the nematophorous Cœlentera. They propose to abandon the division into Hydrozoa and Anthozoa, and to constitute two groups, one to contain the Hydromedusæ which have ectodermal genitalia, the other to contain the Scyphomedusæ and Anthozoa, which have endodermal genitalia, thus denying the supposed close relationship between the two kinds of Medusæ, and making them mere homoplasts, one of the other.

For the two new groups they propose the names Ectocarpa and Endocarpa respectively. Very possibly such a grouping may have to be ultimately adopted. There is no doubt that a sharp line cannot be drawn which shall separate Hydrozoa and Anthozoa, and the new division proposed would not be more wanting in definition than is the present. At the same time, should the grounds put forward by the Hertwigs prove really solid, the old terms proposed by Wilhelm Rapp, of Tubingen, for an exactly similar classification, will have to be adopted, namely, "Exoarii" and "Endoarii."

It is strange that the Hertwigs should have overlooked their countryman's contribution to this subject, published as long ago as 1829, and not so far from Jena ('Ueber die Polypen in Allgemeinen und die Actinien insbesondere,' Weimar). Yet further do the Hertwigs err in their reference to the history of the investigation of Cœlenterate morphology, for, whilst discussing the appropriateness of the old terms, Phanerocarpa and Cryptocarpa, for the designation of the groups which they propose to establish, they refer those well-known terms to Edward Forbes (loc. cit., p. 624) instead of to the illustrious Eschscholtz.

The difficulty which presents itself most obviously in the

way of the proposed new grouping of the *Cœlentera nemato-phora* is found in the Ctenophora. These have been re-cently shown by Haeckel to be most readily explained as a peculiar modification of the Hydromedusæ; at the same time there seems no reason to doubt that their genitalia develop from the endodermal cells of the ctenophoral canals. If this be so, the origin of the genitalia from this or that layer, would prove to be—in the Cœlentera at any rate—a matter of no genealogical import, instead of one of primary signi-ficance.

An important character, serving to unite the *Medusæ acra-spedæ* with the *Anthozoa*, is found by the Hertwigs in the homology of the gastral filaments of the former with the mesenterial filaments of the latter.

Muscular fibre (mesoderm) derived from Endoderm.—For some years the record of observed fact has pointed to the *exclusive* derivation of muscular and fibrous tissues in the Hydrozoa and Actinozoa from the ectoderm, the endoderm being employed solely as an epithelium lining the gastro-vascular system. Recently, however, Professor Claus, who has in recent years by varied and most valuable researches done so much towards placing our knowledge of the Hy-drozoa on a secure histological basis, showed that Halis-temma possesses in its axis a delicate layer of ring muscula-ture derived from Endoderm ('Arbeiten aus d. Zool. Institut der Universitat,' Wien, vol. i, 1878). This observation has been followed by the definite observation on the part of the Hertwigs (loc. cit.), to the effect that in the Anthozoa (Actinia, Cerianthus, &c.) the musculature is chiefly derived from Endoderm, and only a small layer from Ectoderm.

If this observation should prove to be thoroughly well founded, we have a new basis for the interpretation of the facts now known as to the origin of muscular and connec-tive tissues in the higher animals, the Cœlomata or Tri-ploblastica.

Recent Researches on Bacteria.—In a *résumé* which we recently gave (this Journal, vol. xviii, p. 455) of the recent contributions to our knowledge of this most important group of organisms we omitted to make any mention of the important researches of Dr. Alb. Fitz, of the Chemical Institute in the University of Strasburg. These have been published from time to time in the 'Berichte der Deutschen Chemische Gesellschaft zu Berlin' from 1876 up to the present time. Dr. Fitz has investigated from the chemical

point of view the fermentations caused by various Schizo-myceta, and also that due to *Mucor racemosus*. One of his most important results (published in 1876) is that glycerin in the presence of calcic carbonate enters into fermentation by the agency of a Bacterium. The chief products of the fermentation, in addition to carbonic acid and hydrogen, are normal butyl-alcohol and normal butyric acid. As by-products are produced also in very small quantity ethyl-alcohol and one of the higher fatty acids, probably capronic acid. The Bacterium of this particular fermentation has the form of a Bacillus. It is on the average two micromillimeters broad and five or six long. A smaller Bacillus occurs with it in glycerin fermentation, to which is probably due the production of the ethylic alcohol. Dr. Fitz found, moreover, that the remarkable Bacterium which gives the blue green colour to old pus, can also excite fermentation in glycerin. He cultivated this organism from pus by transference to a solution containing calcium lactate and food-salts (ammonium chloride as a nitrogen food). In a successful cultivation he obtained *not* a coloured solution, but a colourless reduction-product of the colouring matter. Only on the surface was there a blue tint. This was due to the access of oxygen to the surface, for when the solution was agitated with atmospheric air, it immediately assumed a deep blue colour, like that of the solution of copper sulphate. This pus-blue colouring matter is soluble in water, is turned red by acids, and by alkalies again blue. It is, therefore, similar to litmus. The Bacteria which produce it, and multiply luxuriantly in the solution, are small elliptic bodies, about one to one and a half micromillimeters in length, and generally occur in couples.

Dr. Fitz's further researches on the fermentation of erythrite, mannite, gelatin, and albumen, as well as those on butyric and propionic fermentation, are of the greatest importance and interest.

The separate tract of Dr. Robert Koch, entitled, ' Untersuchungen über die Ætiologie der Wundinfectionskrankheiten ' (Leipzig, 1878), is worthy of the most careful study from all persons interested in the progress of knowledge relative to the all-important Bacteria. In a first series of experiments we have an investigation of septicæmia produced in mice by injection of putrid blood or infusion of meat beneath the skin. Death in some cases resulted in from four to eight hours, being due *not to the multiplication of Bacteria* but to the poisonous matter (sepsin) produced by them and injected with them. In other cases, death resulted after

fifty hours, the Bacteria (a bacillus form, one micro-millimeter in length) having multiplied in the blood and crowded the vessels of all organs of the body. One-tenth of a drop of the blood of an animal thus affected sufficed to convey the infection, and it was handed on thus for seventeen generations.

A distinct disease—gangrene—was induced in some of the mice experimented on, and this was found to be accompanied by the appearance of micrococci (minute spherical Bacteria occurring in chains and masses). These are, in all probability, a form-phase of the septicæmic Bacilli, which are their necessary predecessors, but the Micrococci differ not only in form but in pathological activity from the Bacilli which generated them.

In rabbits by injection of putrid blood a subcutaneous abscess could be produced without infection of the blood. The disease could be propagated by injections of the fluid of the abscess which contained micrococci. From putrid fluid obtained by macerating a mouse-skin in water, pyæmia was produced in rabbits which could be propagated by injection of $\frac{1}{10}$th of a drop of the blood. Micrococci (not forming chains nor zooglæa masses) characterised this particular kind of pyæmia. Another septicæmic condition was obtained in rabbits by subcutaneous injection of putrid meat, and found to be characterised by large micrococci (one micromillimeter in diameter). This was communicable; but another local erysipelatous condition of the tissues, obtained by injecting mouse-dung infusion beneath the ear of the rabbit, was not inoculable, although the tissues of the ear contained large numbers of Bacilli, three micromillimeters in length.

These investigations are of the highest importance, as showing firstly that a *variety* of inflammatory diseases may take their origin by the access of certain Bacteria to wounds ; secondly, as showing that the *particular form* of Bacterium in each case can be distinguished. The derivation of the micrococcus of necrosis of the mouse from the Bacillus of septicæmia of the same animal is also a result which has great significance.

Dr. Koch is not able to admit in the case of septicæmia that the virulence of the poison increases with each successive inoculation. This he admits only up to the third inoculation as a rule, and attributes it in accordance with the most probable analogy to the history of various beer-ferments studied and separated by Pasteur, to this fact, namely, that just as with beer-yeasts so with septicæmic ferments, cultivation by iterative minimal inoculation in their specially

appropriate media, serves to *purify* the specific ferment from injurious interfering ferments of other species.

M. Paul Bert ('Comptes Rendus Soc. Biologie,' 1879, p. 355) has been led by some experiments on the blood of animals infected with Anthrax (*Bacillus anthracis*) to conclude that such blood may contain two poisonous elements, the one similar in nature to that of cow-pox and glanders, non-organised, and precipitable by alcohol and of a highly virulent character; the other consisting of the micro-parasites known as *Bacillus anthracis*. The first poison which can be obtained free from the second by subjecting Anthrax-blood to compressed oxygen for two days (which destroys the Bacillus) causes death when introduced into a guinea-pig in about twelve hours; the second requires about thirty hours. These results are quite in accordance with the germ-theory of such diseases, and with what is known in general as to the properties of organised ferments. The Bacteria or organised ferments undoubtedly produce poisonous matter, such as the sepsin, now recognised for some years, and above noted by Dr. Koch. Such matter may act directly as a poison or the poisonous effects may be retarded until the Bacterium has multiplied and produced larger quantities of the toxic material.

It is exceedingly probable that soluble ferments are produced in a similar way by many Bacteria associated with fermentative changes such for instance as that of urine.

The Bacteria which *normally* infest the human body must be exceedingly numerous and varied in form and properties. At present they have not been fully recognised. Mr. Butlin ('Proc. Royal Society,' 1879) has made a beginning in describing those which infest the mouth, and which are the chief constituents of the fur of the tongue. Mr. Butlin finds Sarcina, Bacterium, and Spirochæta in the mouth, but chiefly micrococcus and *Bacillus subtilis*.

We have yet to obtain from some careful worker with high power objectives an account of the forms of Bacteria present in the contents of the large intestine and other passages of the body. Further, the different protected surfaces of the skin offer an interesting field for study in relation to this subject. The axillæ, pubes, and even the scalp, have their special saprogenous Bacteria, whilst the epidermis of the prepuce and of the plantar surface of the foot is occupied by Bacteria-ferments, specifically connected with the higher fatty acids.

Publications of the Zoological Institute of the University of Vienna.—We desire to draw the attention of our readers to an important series of memoirs published under the direction of Professor Claus, of Vienna. A very large amount of first-rate zoological research is now being carried on under the direction of the Vienna professor of Zoology. He not only holds his important chair in that city but has the zoological station of Trieste under his direction, and both he and his pupils have made ample use of these fine opportunities. Among the memoirs which have appeared in the 'Arbeiten aus d. Zool. Instit. der Univ.,' Wien, are those of Claus on Halistemma, on Charybodæa, and on Phronima. These memoirs are *not* published in any other periodical or society's proceedings.

New Biological Journal.—On the first of January will appear a new biological journal, published in Brussels. Professor Edouard Van Beneden is one of the editors; the journal will be chiefly devoted to original memoirs and similar in scope to our own. We have great pleasure in commending the new Belgian journal to the attention of histologists and embryologists.

PROCEEDINGS OF SOCIETIES.

Blodgettia.—Dr. E. Perceval Wright exhibited some freshly mounted specimens of *Blodgettia confervoides* of Harvey, which he had prepared from specimens recently collected at Key West by Mr. Farlow, and sent to him preserved in alcohol. These very clearly showed that strings of spores referred to by Harvey were parts of a parasitic plant living in the cells of *Cladophora cæspitosa*. This parasite would seem to consist of a series of delicate threadlike hyphæ, with here and there enlargements. For this Fungoid form Dr. Wright would retain the generic term Blodgettia, of course with a new diagnosis, and he would venture to call the species after Dr. Bornet. In addition Dr. Wright read a letter received that very morning from Dr. Bornet. Alluding to the memorandum about this plant in the Club Minutes for March 23rd, 1876, Dr. Bornet wished to correct the statement referred to there as having been made by him, on the authority of Mr. Farlow, to the effect that Blodgettia was an unicellular form of parasite. It was most obviously nothing of the sort, but a fungus with branching hyphæ.

Coscinodiscus Sol, Wallich, *from Sea of Java, exhibited.*—Rev. E. O'Meara exhibited specimens of the rare diatomaceous form *Coscinodiscus Sol*, Wallich. These were found on slides from material gathered in the Sea of Java and mounted by Professor T. P. Cleve, of Upsala. The form is not enumerated by Professor Cleve among the many new and interesting species discovered by him in gatherings from the Sea of Java.

Anthelia Turneri, Dumortier, *from Kent, exhibited.*—Dr. Moore showed an example in fruit of the very rare British species *Anthelia Turneri*, Dumortier—*Jungermannia Turneri*, Hook., Br. Jung This was first found near Bantry by the late Miss Hutchins in 1811. Dr. Taylor remarks in Flor. Hib. (1836), "This plant has never been seen growing but by its discoverer." Dr. Moore stated he had often searched for it when botanising in the South of Ireland, but never succeeded in finding it. It has, however, been found in France, according to Dumortier in 'Hepat. Europ.' (1874). The specimen now shown came from

North Kent, and was found by H. Davies, Esq., of Brighton, who sent it to Dr. Moore. Like its congeners, this forms a very neat and pretty little object, though the present example had become too dried and brittle to enable one fully to realise its elegance.

Cross Section of the Spine of Acanthaster echinites, Ellis et Solander, was exhibited by Mr. Mackintosh. The genus is remarkable for the possession of numerous madreporic bodies and spines with double articulation, which in section show a pith with rather larger interspaces than are seen in the peripheral part, which is covered externally by a tolerably thick skin. There were no indications of the solid wedges which form so characteristic a feature in the spines of Sea-Urchins.

Problematic cotton-like branched fibrous structure, exhibited. —Mr. Archer drew attention to a problematic piece of organization, met with in an aquatic gathering, resembling a little "tree" of colourless cotton fibre, having enclosed in the stem and branches and running longitudinally therein one or more opaque and shining wire-like fibres, these apparently very brittle, being often here and there seemingly broken into short lengths; the apices of the branches, which proceeded in a tufted manner from the central stem, were apparently granuliferous, the parts lower down hyaline, and more or less flattened and twisted—the question was, what could this thing be?

22nd May, 1879.

Spermatic state of Volvox, showing the groups of spermatozoids and the yellow oospores, were exhibited by Mr. Archer; no meeting of the two elements had been noticed.

Berkleyia.—Dr. E. P. Wright read a letter which he had received from Dr. Grünow in reference to the algal form exhibited by Dr. Wright to the Club in May, 1876. This form, from Dr. Harvey's Friendly Island Collection of Algæ, had been described by Dr. Grünow in the Botany of the Novara Reise as *Berkleyia Harveyana,* which fact had been overlooked by Dr. Wright in 1876. At Dr. Grünow's request he had again carefully and in many ways examined the type-specimens, and he now exhibited mounted preparations thereof, some in glycerine, some simply burnt, and some cleared with weak acid. In none of these was there a trace of silex to be seen, unless here and there an odd stray frustule of a Navicula, a Diatoma, or a Surirella. In addition Dr. Grünow had very kindly forwarded some mounted specimens of his species, of the nature of which there could be no doubt, they being decidedly siliceous frustules, in all probability quite correctly assigned to the genus *Berkleyia* by Dr. Grünow, so that now the question stood thus: *Berkleyia Harveyanus* Grünow, was a diatom from the Friendly Islands. But the

Alga? quam paradoxa! of Harvey was not a synonym thereof, and this latter, a mass of semi-confused congeries of green (phyco-chromaceous?) cells, was still to be described.

Section of Larynx of Human Fœtus, exhibited.—Mr. B. Wills Richardson exhibited with a Quekett's dissecting microscope and one of Charles Baker's 2-inch objectives only, so that the whole of the object might be seen, a cross section of the larynx of a nine months human fœtus.   In the specimen the most striking structures were (1) sections of the thyroid and the arytenoid cartilages; (2) the laryngeal pouches; (3) the pharynx; (4) portions of some of the intrinsic muscles of the larynx; (5) the laryngeal columnar epithelium; and (6) mucous glands. The specimen was a triple staining made with carmine, picric acid, and madder, the latter being an excellent medium for staining cartilage cells, their nuclei and nucleoli.

The section was a striking illustration of the great assistance that may be derived from the use of the freezing microtome in minute anatomical research, for it is difficult to conceive that so even a section could be made without the greatest difficulty by any other methods with which we were familiar.   Moreover, Mr. Richardson observed that scarcely a bit of the larynx was wasted, as he succeeded without any trouble in cutting the whole of it, the tenuity of all the sections being about the same as the one he exhibited.   It was mounted in Klein's dammar solution.

Cross section of the spine of Toxopneustes variegatus (Lamk.) was shown by Mr. Mackintosh, who called attention to the differences noticeable between it and the spine of *T. maculatus*, A. Agass.: these are chiefly the lesser extent of epithelium in the former, the more gradual increase in size of the solid wedges and the marked figure-of-8 shape of the peripheral wedges, those of *T. maculatus* being oval.

Mr. Archer announced that the Royal Microscopical Society of London had done the Club the honour to "nominate it under the bye-law relating to *ex officio* Fellows, and the same having been approved of by a General Meeting, its President for the time being is entitled to the privileges of an *ex officio* Fellow of the Society;" and it was resolved that the marked thanks of this Club be communicated through the Secretary of the Royal Microscopical Society to that body for the distinguished honour thus conferred.

19th June, 1879.

The stated meeting appointed for above evening was not held owing to the very sudden and much-lamented death of Dr. David Moore, F.L.S., M.R.I.A., Director of the Botanic Gardens, Glasnevin, for many years a highly esteemed member of the Club.

July 24th, 1879.

Xanthidium Robinsonianum, Archer, *shown from a new midland Irish locality.*—Mr. Crowe showed examples of *Xanthidium Robinsonianum,* being the fourth occasion this form has as yet been met with; first found by Mr. Archer in co. Armagh, then in co. Donegal, afterwards by Mr. Crowe at Glengariffe, and now in co. Kildare, this well-marked little species has not yet been taken out of Ireland. Although its distribution is thus wide in this island, it must still be accounted as very rare. The form is small, semicells trapezoid, the rounded angles crowned by a few minute spines, central elevation of the semicells bearing a few irregularly disposed, rather conspicuous granules; lower margins straight, isthmus slightly elongated, constriction thus forming a parallel and straight division between the semicells. A more detailed description will be given on another occasion.

Trichia clavata, Persoon, was exhibited by Mr. G. Pim; this was found growing on moss in an orchid basket in his store. This form comes under the genus *Hemiarcyria* of Rostofinski, which differs from *Arcyria* in having the capillitium in spirals, as in *Trichia,* and from *Trichia,* defined by him in having the spiral combined into a dense network similar to *Arcyria.* This proved to be the fully-developed form of the immature myxomycete shown at a previous meeting of the Club. It was abundant on the wood and moss for many weeks. It formed a very pretty object viewed either with a low power, so as to embrace the whole, or with a higher one, so as to exhibit the structure of the capillitium.

Section of Limestone from Ogradina, Banks of the Danube, exhibited.—Professor Hull exhibited a thin slice of limestone from the banks of the Danube, near Ogradina, considered to be of Cretaceous age. The rock from which this was taken rises in massive cliffs above the left bank of that river, where it traverses the Eastern Carpathians. As Dr. Hull could discover no traces of fossils, he had hoped a thin section would probably (as in the case of our chalk) exhibit a foraminifera structure, but this expectation had not been fulfilled, as no organic structures were visible under the microscope in the sections exhibited. The entire field consists of small particles of calcite, which polarise with the usual opalescent colours, and fit into each other along irregular, but generally straight margins. A high power shows the particles of calcite to be cellular in some cases, and to be traversed by cleavage planes in others. Veins of calcite also occur. We must assume the absence of organic structure to be due to the entire replacement of the original rock by foreign matter, but of similar composition, assuming, what is generally admitted, that all limestone formations are of organic origin. The replacement of the original organised structure by crystalline calcite is evidently a subsequent process of "metamorphism," of which the specimen exhibited affords an illustration.

23rd October, 1879.

Peculiarity in Histological Structure of Spine of Echinothrix Turcarum, Peters.—Prof. H. W. Mackintosh exhibited a cross section of the spine of *Echinothrix Turcarum*, Peters, and called attention to a peculiarity in its structure, of which this was, so far, the only example he had met. In the normal spine the solid wedges are coextensive with the network between them, but in this form they were produced beyond it, reminding one somewhat of the arrangement in the spine of *Echinus lividus*. The specimen from which the spine had been taken formed part of a collection made by Professor E. P. Wright in the Seychelles, which had been presented by him to the Museum of Trinity College, Dublin.

A new Unicellular Epiphytic Green Marine Alga, exhibited.— Dr. E. P. Wright showed specimens of a new unicellular epiphytic green marine alga found attached to larger forms in rock-pools at Howth. This was a sub-globose or balloon-shaped, or sub-quadrilateral thick-walled, form often showing empty cells still attached by their lower extremity, their contents evacuated as zoospores. Thus this would be to a great extent like Characium, but the forms there included are stipitate. It called to mind a form found by Mr. Archer some few years ago in moor-pools in Connemara and attached to confervoids, but the cells were cubical, thus presenting a quadrilateral outline and the examples attached (sometimes in rows) by the whole of the base in it, forming the longest diameter.

Roothairs inside the rootsheaths in Azolla pinnata.— Dr. McNab exhibited roots of *Azolla pinnata*, showing the sheath with the numerous root-hairs developed inside the inner sheath, and held down by it with their apices pointing to the apex of the root until they are liberated by the elongation of the root. These hairs are produced in regular whorls close to the growing-point, but by growth of the axis their regularity is disturbed. Strasburger mentions that in Azolla pinnata the sheath breaks up and gives the root a plumose appearance. Nothing like this could be observed either in young roots or in those which have lost the root cup. As Strasburger does not describe any root hairs on these plants, it is probable that they have been overlooked.

Micrasterias furcata, exhibited.—Mr. Archer showed an example of *Micrasterias furcata*, one of the most rare as well as the most beautiful of the Desmidieæ. He had himself seen probably in all not half a dozen examples of this fine species, always from Connemara, and which appears quite distinct from *M. Crux-Melitensis*, itself very rare, but cropping up sometimes, mainly in co. Westmeath gatherings.

Trichia fallax, exhibited.—Mr. Greenwood Pim showed *Trichia fallax*, var. *cerina*, from his stove at Monkstown. This form differs from *T. chrysosderma* in having much finer and more delicate spiral elaters, which are cylindrical and taper abruptly

to a point, while those of *T. chrysosperma* tapered gradually from the middle to each end—the spores being quite smooth in the former and minutely echinulate in the latter.

Zygospore of Xanthidium Robinsonianum, exhibited.—Mr. Crowe, showed the zygospore of this species in continuation of his exhibition of the parent form at last meeting; these he had since discovered in another gathering made on the same occasion. They are orbicular, beset with elongate very slender spines, trifid at apex, slightly dilated at base; the contents of the spore present a somewhat olive-green or bronzy hue, rendering the appearance somewhat marked. It is therefore interesting to have found the zygospore of this species, which is really rare in Ireland, though, as seen, widely diffused, but yet it does not seem to have been met with out of this island.

Nodose Hairs.—Mr. B. Wills Richardson exhibited two mounted slides of human nodose hairs which occurred in the practice of Dr. Walter Smith. The specimens on one of the slides were from the head of a young lady of nineteen, and on the other were from the head of a boy of seven years.

Similar specimens from the girl's head were exhibited by Dr. Smith at the recent meeting of the Medical Association in Cork, and excited much attention. These hairs do not correspond with any of the descriptions of nodose hairs published, at least as far as a very prolonged research could ascertain. Indeed it may be assumed that they are unique, from the fact that Mr. Erasmus Wilson requested Dr. Smith to give him some specimens for the Museum of the Royal College of Surgeons in England; and Dr. Lionel Beale, to whom he (Mr. Richardson) sent a slide of the hairs, wrote to him to say that he had not seen anything of the kind before, and asked for some specimens for the Royal Microscopical Society of London.

The boy's hairs are not so uniformly nodose as the girl's; still his head afforded many perfect specimens, and prettier microscopic objects than hers, being almost altogether free from colouring matter. Many of the imperfect specimens had only a couple of nodes, the remainder of the shaft being normal, There is not the slightest trace of a fungus on or in any of the specimens, neither is there any appearance to lead to the inference that the nodes could have been caused by such. At the constrictions the imbrication of the scales can be easily made out, but only at these points.

The girl's hair, to the naked eye are regularly checked, black and white; the boy's have no such appearances, owing to the absence of pigment. The specimens were mounted in Klein's dammar solution. He might mention before they were examined that those from the boy's had not been previously exhibited.

Tongue of Cyclothurus.—Mr. P. S. Abraham, exhibited a section of the tongue of *Cyclothurus*, the Central American ' Tee Ant-eater,' a microscopical examination of which had not hitherto been made. The section showed the general arrangement of the muscular

bundles, remarkable for the great distinctness with which ' Cohnheim's areas ' came out on the cross section of the fibres. The stratified mucous epithelium was well seen and the submucosa was apparently destitute of mucous glands ; the shiny secretion which lubricates the tongue being probably the product chiefly of the enormous submaxillary glands.

Aorta in Ruminants.—Dr. Purser showed specimens of the aorta of ruminants, cows and sheep. In these animals the muscular tissue of the middle coat does not consist of single film or small groups of films equally distributed between the elastic plates or membranes, but in parts the elastic plates are interrupted and the tissue is composed almost altogether of muscular fibre, with only a few elastic threads interposed. In these muscular patches the fibres run, for the most part, transversely, but in some cases obliquely or longitudinally.

Section of Villous Tumour.—Dr. Charles Ball exhibited some sections of a villous tumour of the rectum, which showed one reason why these growths have been so frequently confounded with true carcinomata, namely, that in consequence of the numerous involutions of the superficial epithelium sections would present the appearance of endogenous growths of epithelium, while in reality these appearances were due to the section having traversed the bottom of superficial sulci.

Encysted State of Vacuolaria virescens, Cienkowski, *exhibited.* —Mr. Archer showed a condition of this conspicuous flagellate, forming in itself a pretty object, and one which, met with in an isolated way, had long puzzled him as to what it could be. Here, however, were examples of this flagellate partially encysted, and others wholly so, the latter still encircled by the original cuticular covering or skin, standing off as a separate pellicular investment. The inner body appeared as a " spore-like " structure, contents dense, green, and quite destitute of any vacuolar appearance, its bounding wall very thick, and covered by a number of large subhemispherical hyaline prominences. This, without the cast-off parent, simply membranous coat, is, as has been said, not unlike a possible zygospore of say some desmidian &c., but something always told one it could scarcely so be. Evidently Cienkowski's original examples had not gone on to the formation of this thickened and unduly margined inner wall, or he would have figured this condition : he represents only a still, smooth, thin-walled condition, not uncommon. Indeed it would only be by obtaining examples in several conditions that the connection between the two extremes could be seen. These bodies, apart from their being still surrounded simply by the parent " cell-wall," could not be regarded as truly "spores" or, as in any way resulting from conjugation ; if they were formed by the union of two individuals, they would have been larger ; on the contrary, they appeared rather smaller, but more dense, than the ordinary freely swimming examples. They

probably merely represent a " resting " or " encysted " condition of this organism.

It was then

Resolved, " That we record with unfeigned sorrow the death of our colleague, Dr. David Moore. The records of the Club testify to the number and value of his scientific contributions; but our members have in addition to mourn the loss of one whom we each and all esteemed as a personal friend, and whose vacant place will leave a serious blank in our limited ranks."

MEMOIRS.

The COFFEE-LEAF DISEASE *of* CEYLON. By W. T. THISELTON
  DYER, M.A., F.L.S., Assistant Director, Royal Gardens,
  Kew. With Plates IX, X, XI, XII, XIII, and XIV.

DURING the last ten years a parasitic fungus has devas-
tated the coffee plantations of Ceylon and Southern India,
and is now slowly spreading eastward through the coffee-
growing countries of the old world. Its history commands
our attention as the enemy of one of the most important
industries of the tropical possessions of the empire. In
itself, moreover, it is an interesting biological study, supply-
ing, as it does, a striking analogy in its first appearance,
progress, and effects, to the epidemic diseases of animals, in
regard to which, however, so far, there is only a presump-
tive probability in favour of the view that the physio-
logical disturbances or organic lesions which are charac-
teristic of them are due to the introduction into their
tissues and subsequent multiplication there of some extraneous
organism.

The early history of the disease is well ascertained in
Ceylon. Before 1869 it was altogether unknown, and the
fungus which causes it had not been detected, notwithstand-
ing that the plants of this group had been diligently collected,
no less than 1190 species (of which two thirds are endemic)
having been described.[1] In May of that year, in a com-
paratively new estate in the Madulsima district, which
occupies the south-western portion of the hilly country, a
few plants were noticed which had light-coloured patches
on the under side of their leaves; and these patches, on
closer examination, were noticed to be dotted over with
orange-coloured dust. In July following two or three acres
were found to be affected in this way. From this time it
rapidly spread, till in 1873 there was probably no estate in
the island entirely free from it.

[1] Berkeley and Broome, ' Journ. Linn. Soc. Bot.,' vol. xiv.

From the first appearance of the disease its serious character was recognised. Specimens of the diseased leaves were sent to this country, and were examined by the Rev. M. J. Berkeley, who named it *Hemileia vastatrix*, and published a short notice describing it in the 'Gardener's Chronicle' for 1869 (p. 1157) with a figure.[1] He not merely determined it to be quite new to science, "but with difficulty referable to any recognised section of fungi." He considered it to be intermediate between *Mucedines* and *Uredinei*, a view which is not sustainable since the Rev. R. Abbay[2] and Mr. Morris, after working upon the plant in a fresh state, have both agreed in affirming that the reproductive bodies regarded by Mr. Berkeley as spores are in reality sporangia. This observation widely alters any possible view of the affinity of the fungus, but it has not, so far, led to any very definite systematic position being assigned to it.

The coffee tree is an evergreen, and the speedy effect of the attack of the fungus upon its young leaves is to cause them to fall off, leaving the tree almost bare. After a time it throws out fresh healthy-looking foliage. Sooner or later this in its turn exhibits the fatal spotting, and is shed prematurely. It is said that there is no known instance of coffee trees being actually killed by the attacks of the disease, but they become much enfeebled by the repeated loss of foliage and eventually become worthless as crop producers.

The following pages give in a condensed form the net result of the investigations to which the life-history of the *Hemileia* has been subjected. The plates have all been drawn from drawings by Mr. Morris, and have been borrowed from a book which he is preparing for the use of the Ceylon planters.

To the naked eye the first appearance of the *Hemileia* is indicated by a slight transparency or "palish discolouration," easily noticed when the leaf is held up to the light. These transparent spots indicate the points where infection of the leaf has begun. As the "spot" becomes larger and older it assumes a faint yellow colour; ultimately on the under side of the leaf it becomes covered with a bright yellow dust, and this later on changes to a bright orange. (Plate IX, fig. 1, represents the under side of a coffee-leaf in a diseased state.) The discoloured patches are irregular

[1] Reproduced in 'Quart. Journ. Mic. Sc.,' 1873, p. 80.
[2] 'Journ. Linn. Soc. Bot.,' vol. xvii, p. 176.

in size, but always preserve a circular outline, except where, as occasionally happens, the accidental coalescence of two patches produces one of irregular form (Pl. IX, fig. 2, A). They are most numerous towards the apex of the leaf, the tissue there being somewhat younger and more succulent, and therefore more easily attacked. The centrifugal development of the patches is sometimes arrested by the barrier presented by the midrib or a vein, but this is not always the case (Pl. IX, fig. 1, A).

When a diseased patch is examined with a low power it is seen to be covered with the somewhat symmetrically disposed clusters and sporangia, the most mature being in the centre (Pl. IX, fig. 4). Further examination shows that these clusters occupy stomata, Pl. IX, fig. 5; and fig. 6 shows a cluster in which some of the uppermost sporangia have fallen off, allowing the oblique attachment of the rest to the mycelial threads which fill up the openings of the stomata to be seen. Mr. Morris describes the fully ripe and detached sporangium as "an ovate, oblong, or reniform body, covered externally on all sides but one with globose or wart-like papillæ. The smooth surface of the sporangium is underneath on the side of attachment, and the fact that on all other sides the sporangium is covered by wart-like papillæ gives it a peculiar character, which, according to Mr. Berkeley, is found in only one other fungus, viz. in *Badhamia*." The immature sporangia are destitute of papillæ.

The *Hemileia* is itself not without its enemies. In Pl. IX, fig. 1 A, a dark-coloured patch is shown where the sporangia have been attacked by some other fungus, possibly an *Aspergillus*. In fig. 3 the sporangia are colourless towards the centre of the spot, their contents having been destroyed by some other agency of the same kind; in this patch also is represented the larva of a small undescribed dipterous insect (fig. 7), which likewise feeds on the ripe sporangia. This larva was first noticed in 1874; Dr. Thwaites then remarked: "Sometimes these little maggots are very numerous, and it could be wished they were still more so, and that thus the fungus spores might be altogether consumed and the propagation of this terrible pest arrested."

If a vertical section be made through one of the diseased patches of a coffee-leaf the connection of the mycelium with the masses of sporangia can be traced and the destructive effect of the former upon the leaf tissue readily studied. The bundle of mycelial filaments, to each of which a sporangium is attached externally, forms a felted mass, like a plug, which fills up the cavity, into which a stoma opens above. This

mass apparently becomes so consolidated that the fact of its composition from compacted mycelium eluded the observation of Mr. Abbay, who thought it was a sac-shaped dilatation of a single mycelial filament.[1] To Mr. Morris is due the credit of clearing up this point, though Mr. Berkeley, in the figure which accompanied his first description, clearly indicated that the sporangia (or, as he thought them, spores) sprang from individual threads.

Tracing the mycelial filaments further into the leaf, which is, of course, reversing the course of their development outwards, they are seen to assume " a branching coral-like habit" (Pl. XII, fig. 2, c). They feed on and destroy the spongy parenchymatous tissue of the leaf where they come in contact with the chlorophyll-containing cells. These are broken down, cavities are produced (Pl. XII, figs. 1 and 2, H), and the leaf at these points appears consequently of a paler colour.

When mature the sporangia appear to be readily detached, and are liable to be dispersed by the wind or by animals, or even the clothes of persons working in or visiting the plantations. Probably the papillæ with which they are covered on all but one side give them a mechanical adhesiveness. At first the sporangia are filled with dark-red or orange granular matter ; when fully ripe they are seen to contain a varying number of minute nucleated spherical sporidia, ranging, apparently, from one or two to fifteen or twenty. According to Mr. Abbay, the sporangia have two coats, an outer one, bearing the papillæ which " disintegrates," while the inner membrane " opens out and slowly dissolves away." Mr. Morris, however, finds that in very ripe sporangia the smooth surface is ruptured, and the sporidia by this means escape. When the sporangia are placed on a moist slip of glass resting on damp blotting paper the sporidia begin to germinate within twenty-four hours, and for the most part inside the sporangium, the mycelial filaments being protruded through its wall, which softens and gives way before it (Pl. X, fig. 2, D); some germinating sporidia, however, are generally found also, which have escaped from the sporangium (fig. 2, c). The result is that everywhere, after an attack of leaf disease, in the neighbourhood of diseased coffee trees, whether on fallen leaves, moist surfaces, or on the stem or branches, a web of mycelium is produced, by the continuous development of which the whole of the exterior of the coffee tree is completely invested with the toils of its

[1] Loc. cit., p. 176.

enemy, although there is nothing to the naked eye which reveals its presence. The extent to which this takes place may be readily judged from the two figures in Plate XI; fig. 1 is a portion of the epidermis of a young branch, and fig. 2 is part of the under surface of a coffee leaf. According to Mr. Abbay there is a mode of propagation in this stage by conidia which are produced during the wet weather which is so favorable to the progress of the mycelial filaments. These conidia are developed at the ends of the branches of the mycelium in the form of radiating necklace-like strings of minute spherical bodies, the whole arrangement closely resembling the fructification of an *Aspergillus*. Whether these conidia are really part of the life-cycle of *Hemileia* must still be regarded with some doubt, inasmuch as fungus-cultivations are peculiarly liable to error from the introduction and development of the spores of other species than that which is the object of study.

Mr. Morris finds that during the four or five months preceding what the planters recognise as " an attack of leaf disease," and which is, in reality, nothing more than the period in the life-history of the *Hemileia* when the sporangia are developed, nearly every part of the coffee tree is invaded by mycelial threads. The absence, at the end of this period, of any empty sporangia leads him to think that the mycelium has travelled from some distance to finally invade the young foliage. If these were infected by sporangia blown on to them by the wind there would be some indication of their presence ; none, however, are found, and it may be concluded, therefore, that the mycelium has travelled to them from the stem and branches, or even from the ground. As this amount of growth could not be sustained by the small quantity of nutrient material contained in an individual sporadium, it must be concluded that the mycelium feeds in its course upon the organic matter dissolved in the moisture which, during the wet season, copiously bedews the sufaces over which the mycelium passes.

As long as the mycelium is external to the coffee plant it apparently does it no harm, however copiously it ramifies upon its surface. But having reached the young foliage during the moist season the filaments begin to invade their internal tissues, and, in fact, to commence the ravages which are so injurious to the health of the coffee tree.

The stomata of the leaves are closely set, and the mycelium is no less closely woven into an interlacing web. Sooner or later, therefore, a filament finds its way into a stoma, or,

passing over it, sends down into its opening a lateral branch.
The mycelial filaments, however derived, now enter the
intercellular air passages of the leaf and immediately alter
their habit of growth. From being slender, filamentous, and
attenuated, they become thicker, more copiously furnished
with short branches, and exhibiting, in fact, what has been
called the "coral-like habit" (Pl. XII, fig. 1, *g*). They
excavate the parenchymatous tissue bounding the inter-
cellular air passages, which is, of course, to attack the plant
in a most vital and important point. The branches may be
seen to impinge on one of the chlorophyll-containing and
starch-forming cells, the wall undergoes absorption, and the
contents are speedily evacuated. These injuries being in-
flicted when the leaf is still young and its new foliage
scarcely matured, the lesion appears to cause a disturbance
of the health of the leaf, at first sight scarcely explicable
by the comparatively small proportion which the tissues
actually destroyed bear to those which are unaffected. But
the internal cavities produced intercept the paths taken
towards the plant by the carbohydrates fabricated by the
leaf, and towards the leaves by the water charged with saline
matter brought up to them from the roots. The functions
of the leaves are completely disorganised, and, as always
happens when this is the case, when the struggle ceases any
longer to be possible, they are shed.

The young shoots which bear the new foliage commonly
perish, and, at any rate, the coffee berries which they bear
are not matured; those on the older part of the branches
are badly developed, and the crop is both reduced and de-
teriorated by the admixture of ill-developed berries. The
effect upon the coffee industry has been very serious. Mr.
Abbay estimates that up to 1871 the average yield for five
years over the whole of Ceylon had been 4·5 cwt. per acre,
whilst for the five succeeding years the average has only
been 2·9 cwt. For the ten years during which the disease
had existed from 1869 to 1878 he estimated the total loss
in crops alone due to the disease as from £12,000,000 to
£15,000,000.

In order to conclude the preceding sketch of the life-
history of the *Hemileia*, it is only necessary to add that, after
feeding for some time on the leaf tissues, the mycelial
filaments send off lateral branches into the air chambers
beneath the stomata, and the aggregation of these grow up
side by side in a closely compacted bundle. On emerging
from the mouth of the stomata each filament develops from
its apex a sporangium. The object of this process of fructi-

fication is apparently to enable the *Hemileia* to bridge over the dry season, the sporidia immediately germinating on the approach of wet weather.

Mr. Morris, from whose forthcoming handbook most of the facts put together in the preceding sketch are taken, was during the greater part of last year placed on special duty for the purpose of coffee-leaf disease inquiry. He was of course extremely anxious to discover some vulnerable point in the life-cycle of the *Hemileia* at which it would be open to attack. It is to his persevering investigations that we owe the clear account of its habits which we now possess. Mr. Morris recommends that during the period of external development, when the mycelium is merely mechanically attached, as it were, to the surface of the coffee tree, it should be treated with some remedy which will destroy its vitality without injuring the coffee tree itself. It is not necessary in the present paper to discuss the attempts and experiments which have been made in this direction, more especially as Mr. Morris will discuss them in his forthcoming publication. But as far as can be judged from experiments on a small scale the destruction of the filaments can be completely effected by dusting the whole coffee plant with a mixture of one part of sublimed sulphur and two parts of lime; the effects are very well shown in Plates XIII and XIV, the former of which shows the epidermis of a branch, and the latter that of the under side of a leaf traversed by *Hemileia* mycelium, both before and after treatment with the mixture. Mr. Morris having been transferred by the Colonial Office from his post as Assistant Director of the Royal Botanical Gardens, Peradeniya, Ceylon, to the Directorship of the Botanical Department of the Island of Jamaica, Mr. H. Marshall Ward has proceeded to Ceylon to take up the inquiry where Mr. Morris left it. There are, of course, wide practical differences between the conditions of a laboratory experiment and the treatment of a plantation. But whatever is the ultimate result, to Mr. Morris belongs the credit of having applied a strictly scientific method of investigation to the problem of coffee-leaf disease, and of having devised the only method of treatment which so far affords any rational hope of success.

It is only fair to observe that Dr. Thwaites, the distinguished late Director of the Peradeniya Gardens, whose opinions must carry great authority, hesitates in accepting Mr. Morris's results. He does not, indeed, gainsay them, but he apparently thinks that the account of the life-history of the *Hemileia* given by Mr. Morris is incomplete, and that the

remedial measures which he has based upon his view of the matter are inadequate.

Dr. Thwaites, in a letter to the Ceylon Government, written September 27th, 1879, points to the fact, which is undoubtedly striking, that the disease, after its first appearance at Madulsima in 1869, showed itself at widely separated points and over considerable areas in the two following years. He thinks this " must, no doubt, have been due to the presence of inconceivably minute germs floating in the atmosphere and carried by the wind." These minute germs are, however, as far as I have been at present able to understand, completely hypothetical, and, as will be seen, it is possible to explain the transference of the disease to remote points from its original centre of dispersion in accordance with known means. Before the investigations of Mr. Abbay had first brought to light the important fact of the extensive development of the *Hemileia* in the mycelial stage, Dr. Thwaites, at a loss to divine how the coffee plants could be infected when the spores had all been shed, hazarded the remark, " In what way the coffee tree receives the infection remains to be ascertained, and from the subtlety of the operation this will have to be inferred, rather than discovered, by direct observation of the process. It would seem most probable that the infecting matter contained in the spore is absorbed by the tender rootlets of the coffee tree, although it is possible to conceive it might also be introduced into the tree through the very young foliage." This was in 1874; in the following year Mr. Abbay, having demonstrated the universal prevalence of the mycelium, Dr. Thwaites felt himself " constrained " to give up the idea of an " absorption of the fungus matter by the roots." He is, however, still unable to account for the fact that after a coffee plant has had all its leaves removed and has been treated with sulphur and lime, its young foliage speedily exhibits once more the evidence of the disease. Mr. Morris attributes this to direct infection by wind from neighbouring plantations or wild coffee trees on which the *Hemileia* is in a state of ripe fructification, and he points out reasonably enough that, till all plantations unite in preventive methods, and wild and neglected coffee is destroyed and rooted out, no method of remedial procedure has a fair trial. Dr. Thwaites, however, in 1874, expressed the opinion that the fungus was " present in the growing tissues generally of the coffee plant in diffused form," and in his latest communication on the subject, dated January 8th of the present year, he states that " the phenomena of the earliest appearance of the leaf

disease . . . . . seem to be very analogous to what are observed in vaccinic or other such contaminations."

Anything falling from Dr. Thwaites on such a subject naturally carries great weight. As has been already remarked, it is possible that the remedial methods suggested by Mr. Morris may prove practically useless. But it may be claimed for them that they are based on intelligible facts, and if they fail the practical proceeding is to try and ascertain why, and improve the curative treatment accordingly. But Dr. Thwaites's criticisms are based, not so much on facts, but on theoretical conceptions. How, it may be asked, can " infecting matter " be absorbed by the roots of a plant to subsequently give origin to a definite organism in its tissues ? How can this organism exist in those tissues " in diffused form ?" And what is the precise meaning to be gathered from these expressions and from the supposed analogy of the *Hemileia* to " vaccinic contamination ?" These quotations are taken from Government documents officially published for the purpose of communicating to the planting community the voice of science on a subject of the gravest importance to them. Dr. Thwaites certainly owes it to himself to express, in a more precise way, and to support by scientific evidence, the views which he has at present only somewhat vaguely indicated, but which seem to lean towards an explanation of the disease as a persistent constitutional infection.

The leaf disease seems to have speedily shown itself in Southern India after its first appearance in Ceylon. In 1876 it appeared in Sumatra, in 1879 in Java and Bencoolen, and this year news has reached this country of its appearance in Fiji, where, however, it was first noticed in May of last year. The outbreak in the latter colony is a misfortune much to be regretted, as coffee promised to be one of its most successful enterprises. Very full details have been sent by its Government to the Colonial Office, and the whole narrative of the outbreak affords a most striking analogy to the introduction of an epidemic, such as that of measles, into Fiji itself.[1]

The disease was in all probability introduced directly from Ceylon to Fiji with a box of seeds packed in earth early in 1879. A few months later it made its appearance in the Great Amalgam Plantation, and subsequently spread to other plantations on the Rewa river, in the apparently capricious manner which Dr. Thwaites found so difficult of comprehension in Ceylon. Fortunately, in Fiji the Government placed the matter in the hands of Dr. W. McGregor, the

[1] Moseley ' Notes by a Naturalist,' p. 341.

Chief Medical Officer, and under his directions a very bold and skilfully conducted attempt has been made to stamp out the disease. His whole report would be well worth printing as a model of procedure based upon strictly scientific principles. The following passages may be quoted :

"On each plantation, the Great Amalgam included, it is easy to see at what point the disease first took hold, and that it is radiating from that spot. The great probability is, therefore, that the disease is carried about from one place to another by physical agencies, a view of its distribution that accords with the glutinous properties [papillose surface] of the spores [sporangia], that cause them to adhere readily to any object with which they are brought into contact. Many people have visited the great Amalgam Estate, curious to see the coffee-leaf disease, and several of them have afterwards visited the other patches of coffee in the same district, and the presumption is that measures were not in each case adopted to prevent the spores of the disease being thus carried from one place to another."

<div align="center">*   *   *   *   *</div>

"On the 22nd I saw the estate of Viti. It was easy to see the disease had there begun on the trees nearest the landing-place from the river, as the trees growing at that spot were covered by the disease for a short distance around them, but the disease became gradually less prevalent as the distance from the landing-place increased, and at the further end of the plantation was not apparent."

The "stamping out" having been sanctioned by the Government, Dr. McGregor gives the following account of the method adopted :

"I proceeded next day to near Viti, the place to be dealt with first, as being the largest and most liable to be traversed by natives and others. As a description of the proceedings undertaken at Viti will give a correct idea of how the same work was carried out at each of the other places, I shall give the former in detail. Our camp was pitched on the other side of the river, nearly opposite Viti. Food was supplied from the native town of Na-Toa-Ika, on the same side of the river as the encampment. In the morning each of the natives employed put aside his sulu, substituting one of banana leaves. I myself and the other two Europeans with me—Mr. Vaughan and Mr. Gibb—took a spare suit of clothes, which we left in the boat by which we all crossed to Viti, and which was anchored in mid-stream during the day when we were at work. At night the men

left their banana sulus on the ground, and were thoroughly washed in the river before they could get near the boat. The Europeans walked into the river, where they and their clothes were thoroughly washed before entering the boat to put on clean clothes. By these means the camp was kept as clean as possible under the circumstances, as this method was rigidly carried out."

The whole of the infected coffee was burned with all the herbage on the ground which could retain the sporangia, and additional fuel was carried on to the ground where necessary, so as to enable a continuous wave of flame to pass over it.

The extraordinary prominence which the *Hemileia* has suddenly assumed is at first sight not a little astonishing; but the ravages of parasitic upon cultivated plants, it is not difficult to see is one of the consequences with which we have to reckon in disturbing the established order of nature. *Hemileia vastatrix* is a parasitic plant which has probably for ages lived in a state of constrained commensalism with some other members of the family of *Rubiaceæ*, to which coffee belongs. The constituents of tropical forests are not gregarious; the native host of the *Hemileia* is, perhaps, some plant of little but botanical interest, of which individuals are scattered at intervals through the forest. Parasitic plants in nature obviously cannot extirpate their hosts, because, if they did so, they would determine their own extinction. They only exist at all, therefore, by doing the minimum of injury to their hosts which is compatible with their own continued existence. But when man places within their reach vast aggregations of plants ready for invasion the latent capacity for mischief of the parasite is brought out to the uttermost, and it runs riot at man's expense. If Ceylon were abandoned to nature the coffee plantations would lapse again into mixed forest; coffee would either succumb to the *Hemileia*, or would only be represented by those individuals which proved able to carry on a commensal existence with it.

It is remarkable that though the coffee districts of the New World are free so far from the *Hemileia*, they are also ravaged by a leaf disease. In this also the parenchyma of the leaf is devoured and destroyed with parallel constitutional effects. The parasite is, however, in this case not a fungus but the larvæ of a minute moth (*Cemiostoma coffeellum*). This seems to have first taken to its mischievous vocation in the West Indian Islands, and thence to have been introduced on the South American continent, in all the coffee-growing districts of which it is a serious pest.

On SHEPHEARDELLA, *an* UNDESCRIBED TYPE *of* MARINE
RHIZOPODA; *with a* FEW OBSERVATIONS *on* LIEBERKÜHNIA.
By J. D. SIDDALL. With Plates XV and XVI.

A FRESH interest has attached to the Rhizopoda of recent
years, as extended research has furnished data for the more
systematic study of the group. But the labours of Hertwig,
Cienkowski, Archer, F. E. Schulze, and other eminent histo-
logists, so admirably summarised by Professor Allman in his
two Presidential addresses to the Linnean Society, seem
rather to suggest how much there must be still to learn re-
specting the life-history of sarcode organisms, than to render
us satisfied with what is already known concerning them.
Under these circumstances it has appeared to me that the
following notes on certain little understood forms, especially
on one that does not appear to have fallen under the notice
of previous observers, may be worth placing on record.

On the 22nd of August, 1879, I received from my friend
Mr. Shepheard two bottles of sea-water containing Polyzoa,
Hydrozoa, and Sponges, which had been collected at low tide
on the previous day from the shore near the Lydstep Caverns,
Tenby. For their better examination I placed a number of
the specimens in cells, and had the pleasure of finding that
most of them were living, notwithstanding their long railway
journey in small bottles. On the following day my atten-
tion was arrested by an organism, apparently a Rhizopod,
which had separated itself from the other things in the cell,
and was adhering to the glass, and which, but for the pseudo-
podia extended from its two extremities, would probably
have been passed over as a worm. With a view to the closer
study of this creature, a further supply of material from the
same locality was subsequently obtained for me by Mr.
F. Walker, of Tenby, who, during the months of September
and October, made various gatherings, from each of which I
obtained specimens not only of the species originally found,
but also of one or two other Rhizopods having features of
interest.

To Mr. Shepheard and Mr. Walker, therefore, I am pri-
marily indebted for the material from which the present paper
was written; and, as the organism I purpose in the first place
to describe appears to be new, I have much pleasure in
associating it with the name of my friend and fellow-worker
Mr. Shepheard.

SHEPHEARDELLA TÆNIFORMIS.—*Nov. Gen. et sp.*

General description.—The body of the Rhizopod is uni-cellular, elongated, and abruptly pointed at both ends. Flat-tened and ribbon-like when in a state of activity, rounded and worm-like when at rest. It is furnished with a flexible, transparent, colourless integument of considerable firmness; and the whole tubular cavity is densely filled with yellowish, coarsely granular protoplasm, having a very distinct oval nucleus, and occasionally also a few scattered non-contractile hyaline vesicles. The sarcode rotates in a regular stream around the interior of the integument, and carries the nucleus along with it; the current performing a complete circuit within the cell. The nucleus is seldom carried entirely round the cell, being as a rule intercepted in its course along one stream and passed over into the opposing one before it has travelled far from the centre in either directions. The opposing streams of sarcode thus formed on the two sides of the cell, are not separated from each other by any clear line as in *Characeæ*, though otherwise much resembling the pheno-menon observable in the living vegetable cell. The prin-cipal difference between the two is in the direction of the current, that of *Chara* advancing in a spiral direction, whilst that of *Shepheardella* completes its circuit in one plane, the two currents slightly overlapping each other. The in-tegument is perforated at each end by a minute aperture, through which some of the sarcode passes and collects into a small mass. From this mass a very delicate coating spreads over the whole exterior surface of the integument, and this thin layer occasionally throws out a pseudopodium. But the great network of inosculating and branching pseudopodia are at the two extremities of the organism, extending them-selves from the terminal masses of sarcode to a distance considerably exceeding the whole length of the body, as in Pl. XV, fig. 2. The circulation of the finer granular sar-code in the pseudopodia is easily traced. It is very rapid, advancing and returning in single, double, or even triple streams, according to the breadth of the pseudopodium traversed. So that from the rotation of the chief mass of sarcode within the test and the external circulation in the pseudopodia, it is evident that every part of the body-contents, the nucleus excepted, is in turn brought in contact with the surrounding water. The combined movements present an evidence of vigorous life rarely exhibited among the Rhizopoda, and the transparency of the "test" permits the internal functions of the organism to be as easily followed as the external.

Size.—The first specimen observed measured in length of body, without the pseudopodia, ·07 inch (1·75 millim.), and in breadth ·0016 inch (0·042 millim.). Since then I have measured a number of others, the largest of which is ·31 inch (7·5 millim.) in length, and ·2 inch (0·5 millim.) in breadth. The rest vary between these two extremes. (Pl. XV, fig. 1.)

Two days after finding the first I observed a second much larger specimen, and examined it carefully whilst living with powers varying from 280 to 800 diameters ($\frac{1}{4}$ in. objective with A to D eye-pieces). Nothing fresh was noticed in the sarcode, except that it contained, in addition to the dark granules of irregular size and shape, a considerable number of spherical and oval masses, relatively larger in size, of clear colourless protoplasm. These were carried along by the rotating current, but did not travel so quickly as the finer portion of the sarcode, probably owing to their greater size and weight. No traces of a contractile vesicle could be seen although carefully sought for. Neither structure nor surface markings of any sort could be discerned in or upon the integument, which presented a perfectly homogeneous texture, and may therefore be regarded as a simple layer of hardened or otherwise slightly modified protoplasm.

The nucleus proved to be a far more difficult subject for study, but careful observation in upwards of twenty examples has within the last few weeks revealed its structure. Examined in the living state very little can be made out satisfactorily. In appearance it is a simple oval sac or cell possessing a wall of exceeding tenuity, and filled probably with fluid of greater transparency than the sarcode which surrounds it. The chief difficulty arose from certain dark lines, continually travelling over its surface; not ceasing either when the nucleus was carried along with the rotating sarcode, or momentarily arrested. Some of the varying appearances thus presented are shown in Pl. XV, fig 4. After watching these lines for many hours I was led to attribute them to alternate wrinkling and distension of a nucleus-membrane, taking place without any regularity as to time. They appeared generally to open out from the centre to the edges of its longer diameter, or *vice versâ*. This seemed to prove definitely the existence of a distinct wall of greater or less resistance coating the nucleus, and it appeared to me that whilst the lines were chiefly the result of the unequal external pressure of the impinging currents of sarcode, they might also be in some degree due to inherent causes; and continued observation has led me to believe that the latter rather than the former is the correct explanation.

On the 12th of March last I noticed a fine *Shepheardella* entangled among the tentacles of a " Hydratuba," and placed it between two thin cover-glasses for examination with higher powers, care being taken not to press it. Urticating threads of the lasso-cells had pierced the integument of the rhizopod in many parts, but did not seem to cause any inconvenience. The nucleus was finely developed, and was watched most closely, in order to determine, if possible, whether its movements were the same, or an effect of the rotation of the sarcode. These movements proved, however, more puzzling and erratic than ever, the difficulty being increased by the density of the sarcode. Finding that no satisfactory results could be obtained otherwise, I subjected the creature to sufficient gentle pressure to force out most of the sarcode, when I had the satisfaction of observing that the nucleus continued its meandering course, somewhat after the fashion of a large Paramæcium, for about half an hour after all motion had ceased in the sarcode which surrounded it. The rotation of the sarcode was at once interrupted and speedily stopped altogether by the pressure applied ; the nucleus gradually ceased to move to and fro in about thirty minutes afterwards, a few signs of life being exhibited by its protoplasmic contents for a few minutes longer. The nucleus membrane was deeply contricted and wrinkled into a constantly varying series of elevations and depressions, forming avenues, through which the granular sarcode passed, and at times appeared to receive increased impetus in doing so. In consequence of these ever-changing appearances the various parts of the nucleus could only be distinguished from each other at rare intervals. Sufficient, however, had been seen to prove that the nucleus possesses the power of independent movement, but probably not in so high a degree as the sarcode, in the more powerful stream of which it is, as already stated, carried along with irresistible force.

With two exceptions the specimens of *Shepheardella* examined have each possessed one nucleus. One of the exceptional specimens was rather small and pale coloured, and contained no visible nucleus ; the other was of full size and colour, and possessed three nuclei, all of which were of full size and exhibited the same phenomena of moving lines. They were carried along independently in the sarcode stream, sometimes coming into collision and becoming blocked for a short time, and then forced on again with a jerk by the collected force of the choked current.

This particular specimen was first observed on the side of a bottle, to which it adhered so firmly by its extended pseu-

dopodia, that it was necessary to use a camel-hair pencil to transfer it to a cell. In so doing I twisted it twice in the course of its length, thus causing stoppages of the sarcode-current. Before long, by a series of spasmodic movements of its body, the creature succeeded in stretching itself out quite straight upon the glass, when the jerking movement at once ceased. As the internal rotation resumed its course, slowly at first, and then more quickly, pseudopodia were emitted plentifully. Since then I have observed the same irritability in several other specimens, and believe that the source of such movements is the nucleus, which furnishes the impetus required to produce them. I have never before seen any Rhizopod exhibit the same sort of irritability, or make such manifest attempts to rectify itself when placed in an abnormal position. The power of curving laterally, combined with that of crawling by the aid of its pseudopodia, enables *Shepheardella* to move from place to place with considerable rapidity ; but its habit is, when once fixed, to remain for many days in the same spot with its pseudopodia fully extended.

Abnormal forms.—Several specimens observed during the months of October to December differed considerably from the normal form and also from each other. Two of these are represented in Pl. XV, figs. 5 and 6. For some time I looked upon them as distinct species, but having since traced other specimens of *Shepheardella* through similar conditions during their "breaking up," I am disposed to consider them as representing an early stage in that process. One of them (fig. 6) was found on Nov. 29th. Its body was continually changing in form, pseudopodia being extended from no less than thirteen distinct orifices in its integument ; some of the main branches of pseudopodia reaching to the extraordinary length of three-fourths of an inch (20 millim.), and capturing infusoria in all directions. Most of the food so captured was digested outside the body, in temporary masses formed by the accumulation of sarcode from the pseudopodia. The bodies of the abnormal forms were also full of dark-coloured food particles, diatoms being occasionally noticeable among them, but the sarcode of the normal *Shepheardella* seldom seemed to contain any alimentary or other foreign matter. On one occasion I detected a typical specimen in the act of conveying towards an aperture a relatively large mass of apparently fæcal matter, so large that it choked the orifice for a considerable time and was eventually ejected suddenly and passed away by an excurrent pseudopodium, and finally thrown off at some distance from

the body. The chief food of the animal appears to consist of Infusoria, which are not as a rule conveyed into the body, but digested among the pseudopodia outside the test.

Life history.—The phenomena connected with the life history and reproduction of this remarkable Rhizopod have not yet been fully made out, so that the remarks now offered are in some measure provisional, and subject to correction at some future time. They may be given chronologically, beginning with a note made August 27th, when two *Shepheardellæ* under observation in a cell disappeared entirely during that day and the following night, leaving as their only traces a very great number of Amœbæ of various forms, some of which are reproduced in Pl. XV, figs. 16, 17, 18. What had become of the integuments and coloured granular portion of the sarcode I failed to find out. All the Amœbæ were quite colourless ; most of them possessed a nucleus, or more than one, and a contractile vesicle. They were very active in their movements for from four to six weeks, increasing considerably in size during the earlier portion of the period ; but towards the end of that time their movements became slower and more feeble, and finally ceased altogether, when they collapsed into motionless specks of very finely granulated sarcode (Pl. XV, fig. 19), ultimately becoming so indistinct as to be quite unrecognisable amongst the organic *débris* with which the cell had got somewhat obscured.

Another point noticed was that if some of the sarcode from the extended pseudopodia of a *Shepheardella* was taken up on the point of a camel-hair pencil, and broken up into a number of particles in a drop of sea water on a glass slide, each separate particle presently put out long filamentous quivering pseudopodia ; and if pseudopodia from two such particles happened to touch each other they at once coalesced. After a very short time these characteristic pseudopodia were all retracted, and the particles became quiescent, presently resuming active life, not as before, but as Amœbæ, crawling by *lobose* prolongations.

With a view to ascertaining if possible the mode of reproduction, I put two fine *Shepheardellæ* into a cell on December 9th, and on the 10th, found they were alive and in all respects similar to Pl. XV, fig. 2. By December 15th, at 7 p.m., one of these had assumed the nearly spherical shape of Pl. XV, figs. 9, *a, b, but was still encased by its integument.* Two and a half hours later it had stretched out again, and become in form nearly like Pl. XV, fig. 6, pseudopodia being put out from ten different orifices. The changes of form during the

ensuing two hours were very considerable. By the follow-
ing morning it had again become nearly spherical, and
showed a tendency to divide into four parts by constriction.
On the 17th, at 9 a.m., all the contents were found to have
been expelled from the test during the preceding night, and
the now naked sarcode had divided into four unequal por-
tions (Pl. XV, fig. 10), the empty, cast-off integument lying
near to them in a wrinkled mass. The four separated por-
tions changed shape very slightly and slowly, delicate blunt
extensions of transparent protoplasm occasionally protrud-
ing from the edges of each. In the evening of the same day
rapid changes were continually going on ; the sarcode passing
in *sheets* from one portion to the other. The drawing at
Pl. XV, fig. 11, gives a good general idea of the phenomena at
this stage, though the actual condition varied every instant.
The principal masses literally poured their sarcode one into
the other. New centres towards which all would flow were
formed every few minutes. Some of these for a short time
would form separate and distinct masses, and then reunite
by the coalescing of the sarcode extensions ; so that there
was sometimes but a single mass, which within a few
minutes might be broken up into half a dozen. These
alterations continued for several hours, during which time
no trace of a nucleus, which had become invisible on the
first constriction of the Rhizopod, could be found. On the
following morning, December 18th, 9 a.m., the whole had con-
tracted into one principal mass (from which both lobose and
filamentous pseudopodia were freely extended) except some
minute detached patches of the sarcode which were lying at
some distance from it. These exhibited feeble amœboid
movements so long as they remained free, but were at once
reabsorbed into the principal mass on coming in contact
with any of its pseudopodia. Examined again in the even-
ing of the same day it was found in the same condition, but
a still larger number of the outlying amœboid particles of
sarcode had during the interval become detached. One
very small spheroidal particle (Pl. XV, figs. 15, *a*, *b*), contain-
ing rather larger granules than those usually present in the
sarcode, was removed to a considerable distance, and care-
fully watched until January 25th, by which time it had
gradually given off all its contents, and apparently melted
away. By December 18th, a very large number of Amœbæ,
differing in no recognisable way from the ordinary forms, and
evidently separated from the sarcode which had been con-
stantly in motion for the past nine days, were travelling
about all over the cell cover. These, when once fairly

established and differentiated into endosarc and ectosarc, crawled with impunity into actual contact with the pseudopodia of the other *Shepheardella*, which had up to this date retained its normal form, without in any one instance becoming absorbed, as was the case in their earlier stages.

During the night preceding December 18th the second specimen began to change its form. Its appearance at 9 a.m. on that day is represented in Pl. XV, fig. 7. The sarcode had entirely left one portion of the integument, and collected at the other extremity, which was much swollen in consequence. The nucleus could still be seen clearly, and the internal rotation went on in the sarcode, but, instead of two apertures, it now possessed at least four, from each of which pseudopodia were emitted. The contraction of the sarcode continued slowly and steadily throughout the whole of this day and the next, the nucleus became more obscure as the protoplasm became denser, and finally was lost to view altogether, the pseudopodia being at the same time gradually withdrawn from the wrinkled end. At 7 p.m. on the 19th the now much altered *Shepheardella* adhered to the cell by a few pseudopodia from one end only, and had become broadly oval in contour, as in Pl. XV, fig. 8, *a*, *b*. Examined twenty-four hours later, the integument had become completely emptied and lay in a wrinkled mass on the free end of the exuded sarcode (Pl. XV, fig. 9, *b*, "*i*"). The naked body was adherent to the glass by its opposite extremity, and was deeply constricted in a quadripartite manner, and evidently about to subdivide. Further observations on this specimen were unfortunately prevented by the partial leaking of the cell and the consequent loss of the object.

To return now to the first specimen, whose changes up to the evening of December 18th have been already noted. I found on the 19th, at 9 a.m., that it had once more divided into two subglobular masses, which continued to give out pseudopodia freely, especially under the influence of the warmth and light of a lamp, until the 27th, when the only remaining evidence of life observable in them was a feeble swarming among the granules of the sarcode, and this also ceased after a day or two. After remaining motionless from December 29th until February 15th one of the masses suddenly gave evidence of life by emitting, whilst under observation, a lobose extension of protoplasm containing three enlarged granules. In a short time this travelled away as an Amœba, within whose body the granules appeared as nuclei, no vesicle or vacuole being then present, but, examined again next day, the granules were found to be re-

placed by one very large hyaline vesicle containing denser matters within it. Watched for some time this vesicle was seen to travel towards and ultimately protrude beyond the protoplasm of the Amœba, then rupture and discharge its contents into the surrounding water, wherein they appeared as bright spots held together by a viscid substance, the vesicle afterwards collapsing and disappearing entirely. From one of the two masses a thin layer of protoplasm is still (March, 1880) extended, immersed in which are a number of very transparent and much enlarged nucleated granules, of tolerably regular form. In the larger of these a general slow movement of the contents is discernible, and some which became detached afterwards put out *Actinophrys*-like pseudopodia, as represented in Pl. XV, fig. 13, *a, b, c*. These new forms generally develop a nucleus and small internal contractile vesicle, and in one instance a specimen divided into three distinct individuals. Like the Amœbæ, most of these have now become quiescent, and the two masses of sarcode have become smaller and paler in colour, in consequence of the detachment of particles from them.

From the foregoing account it will be gathered that little beyond the dissolution of *Shepheardella* into amœboid particles has yet been quite satisfactorily traced. No attempt at fission, encystation, or anything approaching to either, and no development of special reproductive bodies, unless we accept as such the minute spheroid drawn on Pl. XV, fig. 15, having yet been observed. The loss of the specimen possessing three nuclei was a matter of much regret, as I had hoped, judging from what has been noticed in other simple organisms having more than one nucleus, that it might ultimately divide into three distinct individuals, and by so doing give conclusive evidence of at least one process of reproduction. As it at present stands, the life history of *Shepheardella* may be looked upon as a chain, a few links of which are here presented, the major portion being still missing.

The similarity of *Shepheardella*, in point of external form, with the *Gregarina gigantea* of M. Van Beneden, has attracted my attention ; but as it is not known that the *Gregarinidæ*, except in the very earliest stages of their existence, resemble the Rhizopoda in possessing the power of extending pseudopodia, the idea which presented itself that the animal I have described might possibly be only one stage of some large parasitic creature of this class, has not yet been seriously followed up. Should there be anything in its life history to verify or controvert this superficial resemblance

it will doubtless become apparent in continued observations.

Histology.—Beyond what has been stated nothing has been elicited from living specimens concerning the structure of either integument, sarcode, or nucleus, but the examination of a series of mounted examples, some of which had been previously treated with reagents, has revealed some further interesting details in the two portions last named.

As to the integument, further investigation only corroborates the description already given, that it is a perfectly transparent, structureless, homogeneous membrane, flexible and elastic. Viewed in optical section with high powers, that is to say magnified from 600 to 1000 diameters, it appears to be double under some conditions of illumination; but this is probably an optical illusion, caused by the unequal density of the surfaces, for when torn across it does not exhibit a double structure, nor is it separated into distinct layers by the action of reagents. Dilute acetic acid softens and swells it, and iodine stains it brown, but it is unaffected by carmine.

Lying immediately within the integument, and closely adhering to it, is an exceedingly thin stratum of very finely granulated colourless protoplasm. Acetic acid causes this subcutaneous layer to rise into clear spherical masses destitute of granules but furnished with a large number of vacuoles. A camera tracing of it, mounted in glycerine jelly, magnified 600 diameters, is given on Pl. XVI, fig. 1. The sketch is taken from a specimen in which the denser sarcode occupying the interior had retreated naturally from one end, leaving the integument with this delicate lining layer exposed. The nucleated granules and masses of firmer protoplasm seen in the sketch are detached portions of the internal sarcode remaining upon it.

The densely granular yellowish sarcode which occupies the whole interior of the body is affected in a very marked manner by osmic acid, dilute alcohol, and picro-carmine, the use of which was suggested to me by Professor Lankester. By the application of these reagents each granule is rendered distinct and separate, and has the appearance of a nucleated cell, the central speck taking the carmine colour, whilst the peripheral portions remain yellow. The drawing (Pl. XVI, fig. 2) is a camera tracing of a portion so treated, mounted in glycerine, and magnified 600 diameters. Some of the granules exhibit different stages of multiplication into two or more by fissiparous division, and their minute size will be realised by the figure referred to.

The number of these nucleated granules contained in each perfect *Shepheardella* is, of course, very great. They are packed so closely that the sarcode seems to owe its density and yellow colour to their presence, and yet it is not difficult to believe that each one of them may, by the continued exercise of the process of free-cell formation, and possibly, also, the exudation at the same time from each cell of a coating of living protoplasm, possess the power of becoming, under favorable circumstances, a new individual like the parent *Shepheardella*. The probability of this is increased by the great enlargement in size which takes place before the granules are liberated as separate living organisms, as shown in Pl. XV, fig. 13 *b*.

The nucleus, which in living specimens appears to be so simple, is found by the more deliberate examination of mounted specimens to be of exceedingly complex structure. It consists of three portions, viz., a lenticular oval body, which may be termed the nucleus proper, an embracing but not continuous inner coat, and an outer membranous sac enclosing the whole. The nucleus proper is an oval biconvex cell, bounded by a very delicate wall and filled with protoplasm, which in Pl. XVI, fig. 5 *b* and *a* will be seen to be contracted and coagulated by the action of the reagents round the edges of the cell ; and at fig. 4 *a* to be dispersed over its whole interior surface. It possesses within it a distinct secondary nucleus of somewhat denser protoplasm, and in this again a central spot or nucleolus of its own. Of the two surrounding envelopes, the inner one is a simple transparent membrane very slightly roughened or granulated in texture, apparently composed of firmly coherent specialised protoplasm, and on this the nucleus lies ; the free thickened edges of the envelope enfolding but not completely covering it (see Pl. XVI, figs. 3 *b*, 4 *b*, 5 *c*, 6 *c*). The outer envelope is a simple membranous continuous sac, completely enclosing both nucleus proper and inner coat (Pl. XVI, figs. 3 *c*, 5 *d*, and 6 *d*). It is perfectly structureless, and so transparent that when tightly stretched by the widely opened inner coat it cannot readily be distinguished, but is easily seen as a somewhat wrinkled thin skin when the inner coat is enfolded.

Figures 4 and 5 are taken from specimens treated with dilute acetic acid and subsequently stained with carmine solution. This method of treatment appears to answer better than any other for rendering the structure clear, and at the same time staining the nucleus proper and its contents, whilst the coverings retain pretty much their original colour. Picro-carmine, after osmic acid and dilute alcohol, renders the nucleus generally very distinct, staining it red, while

the sarcode remains yellow; but it has not so good an an effect in other respects. Figures 3 and 6 are from selected specimens mounted like the last two in glycerine jelly, but not previously treated with reagents. In the former the nucleus proper is plainly discernible, but its coverings are somewhat confused, whilst fig. 6 shows the two coats very clearly but not the nucleus itself.

From the appearances that have been described, it is evident that the moving lines noticed in the nuclei of the living specimens are caused by the alternate opening and closing of the folds of the inner envelope, and that the lines indicate the approximation and separation of its free thickened edges. This movement is probably initiated by the contraction and dilation of the nucleus proper, thence communicated to the embracing envelope, and by it rendered apparent.

The object of this movement, which takes place without any regularity as to time, it is not easy to conjecture. It does not appear to have any very close connection with, or effect upon, the rotation of the sarcode within the body of the animal; but that it serves some definite purpose, probably an important one in the economy of the creature, can scarcely be doubted.

All the sketches of the nuclei were drawn originally twice the size of the engraved figures, as it was found that the details of structure were better displayed under higher magnifying power.

At present it would be premature to assign to *Shepheardella tæniformis* any systematic position, nor could this be attempted satisfactorily until the particulars of its life history are more fully known. What has been already made out appears to indicate that it belongs to the Rhizopoda, but there seems no type hitherto described with which it has any close affinity. Apart from its remarkable nucleus, there would be little reason why it should not be placed near *Lieberkühnia Wageneri*, in company with which it occurs at Tenby; but the presence of the complex body, the structure and movements of which have been described, suggests a somewhat higher organization than we are accustomed to associate with Rhizopod life. Upon this point, as well as upon others which have been mentioned, I trust to be able before long to add to the notes now offered, which must be regarded as tentative only.

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The other Rhizopod to which I have referred, *Lieberkühnia Wageneri*, was first found in the neighbourhood of Berlin by MM. Claparède and Lachmann, and was described and

figured in their ' Études sur les Infusoires et les Rhizopodes.'[1]
The original figure (reproduced by Dr. Carpenter in his
' Introduction to the Study of Foraminifera,' Pl. II) is a
lateral view only. I am now able to give some details in
respect to the mouth, as well as concerning other points in
its structure, from specimens obtained from the Welsh
coast.

It is singular that so remarkable an organism does not
appear to have been observed, except by its original dis-
coverers, until found by me in 1878. My specimens were
obtained from the sides of a bottle of sea-water, in which
I was keeping some Hydrozoa and Polyzoa collected
on the shore, at low water-mark, near the Little Orme's
Head, Colwyn Bay. It may be also of some interest
to observe that the same gathering furnished fine specimens
of *Haliphysema Tumanowiczii*. Its non-appearance for so
long an interval may be accounted for by the fact that in
English books it is invariably stated to have been originally
found in " unknown *fresh* water from Berlin ;" but having
found it in 1878 in sea-water from Colwyn, and subsequently
in sea-water from Tenby, either the Berlin habitat must be
erroneous, or the creature lives indifferently in marine and
fresh water, the former being the likeliest hypothesis. If
vessels containing marine Algæ and other organisms, which
have remained undisturbed for a few weeks in the autumn
of each year, were carefully examined, both *Lieberkühnia*
and *Shepheardella* would probably be found to be not un-
common British species.

In general contour *Lieberkühnia* is ovate or rounded, but
the outline is constantly slowly changing by the movement
of the enclosed sarcode. The actual change in shape is not
very great, and consists principally in the gradual dilation of
one part or other of its circumference, causing it to vary
from an elongated oval or pyriform to a subglobular form.
The milky-white, semitransparent, coarsely granular proto-
plasm, of which the body consists, is invested by a thin pel-
lucid membranous integument, the interior surface of which is
lined by a transparent " subcutaneous layer " of finely granu-
lated protoplasm. Its exterior surface is set with a number of
highly refractive, short, rod-like spicules, disposed at various
angles upon it, like those described by Greeff in *Heliophrys
variabilis*. These spicules are indistinguishable in the living or-
ganism, being covered by the coat of motile protoplasm, but are
brought into view when this is absent, as in dead specimens.
The investment is sufficiently flexible to follow closely all

[1] Geneva, 1850—1861.

the changes in contour of the sarcode mass, and therefore not immediately apparent in the living animal, but is occasionally exhibited very plainly by the contraction of the sarcode within it (Pl. XVI, fig. 8). The sarcode rotates constantly within this integument, and emits at the same time from a terminal orifice a main stem of protoplasm, from which the freely inosculating and branching pseudopodia principally arise. Pseudopodia are also numerously put forth by the coat of living sarcode which, spreading from the main aperture, completely invests the integument. No distinct nucleus, such as that of *Shepheardella*, can be detected in the body, but there are several clear spaces, as shown in fig. 8. These are very distinct, as they are carried along by the rotating protoplasm, and are persistent in form and size, partaking more of the nature of vacuoles than contractile vesicles. They do not appear to possess any wall, and are not seen in a mounted specimen. In addition to these vacuoles, a large number of " vesicular nuclei" are dispersed throughout the sarcode, consisting apparently of a simple cell enclosing a clear fluid and one or more nucleoli. These are especially distinct in a specimen mounted in glycerine, a camera tracing of a portion of which is given on Pl. XVI, fig. 5, showing in optical section the integument with its spicula (*a*), the vesicular nuclei (*b*), the granular protoplasm (*c*), and the " subcutaneous layer" (*d*).

The " vesicular nuclei" are apparently identical with the bodies so designated by Prof. Ray Lankester in his description of the sarcode of *Haliphysema Tumanowiczii*.[1] They are probably not quite so large in *Lieberkühnia*, in which their average diameter is about $\frac{1}{2000}$th of an inch (0·013 mm.), the size of those present in *Haliphysema* being about $\frac{1}{1700}$th of an inch. They seem to be equally abundant in both cases, but there appears to be some difference in the manner of their distribution throughout the sarcode. Prof. Lankester having observed that those present in *Haliphysema* were "scattered in the protoplasm, ... being most abundant in the basal portion of the core," whereas in *Lieberkühnia* they are principally found resting *on* the outer surface of the internal protoplasm, just within the subcutaneous layer. I have not been able to detect as such any of the " egg-like bodies" of the same author, but as the only specimen I at present have is mounted entire in glycerine jelly without previous treatment by reagents, they may possibly be there, although indistinguishable from the nuclei.

This apparent identity of the structure of the sarcode of

[1] 'Quarterly Journal of Microscopical Science,' October, 1879.

Lieberkühnia with that of *Haliphysema* appears to me interesting, as corroborative of the conclusions suggested by Prof. Lankester's paper, viz. that *Haliphysema* is correctly placed among "arenaceous" *Foraminifera*. The affinity existing between these two forms is further demonstrated by the scattered spiculiform bodies with which the integument of *Lieberkühnia* is invested.

The orifice or mouth of *Lieberkühnia*, as seen when the creature was crawling mouth upwards on the thin glass cover of a cell, is situated at its broader extremity in a depression formed by the infolding of the four lobes of the integument, taking the contour of a square whose angles are carried outwards into projecting points (Pl. XVI, fig. 9). The sarcode stolon is shown in the sketch as a small oval spot in the centre of the square—the pseudopodia which arise from it having been omitted to avoid confusion.

To the life history of *Lieberkühnia* I can add little to what has been already recorded, but the following may not be devoid of interest as illustrating its voracity and its general appearance whilst feeding.

On the 5th of October last I found a very fine specimen anchored to the side of a bottle, and transferred it to a cell by means of a camel-hair pencil. When examined under the microscope it was found to be in the act of trying to swallow a living worm quite twice as long as itself, which it had evidently seized whilst in the bottle, retaining its hold upon it even when taken up by the brush for transfer to the cell. The means adopted to accomplish deglutition were somewhat singular. Having got one end into its mouth, the *Lieberkühnia* passed a thick stream of sarcode along the worm to its furthest extremity, the creature meanwhile making feeble efforts to escape from the glairy mass. Then, having got well hold of its prey, and anchored itself to the glass by an outspread network of pseudopodia, it proceeded slowly and deliberately to envelop it, partly by retracting the thick band of sarcode, and partly by advancing its own body. This process continued until the pointed end of the worm was pressed so tightly against the lower extremity of the *Lieberkühnia* as to almost burst the integument, when, finding it could engulf it no further, more sarcode flowed from the interior of the Rhizopod over the worm, until it was completely enclosed (Pl. XVI, figs. 10 and 11). Matters remained thus for some time, the rotation of the sarcode going on with unusual rapidity meanwhile, and the pseudopodia being spasmodically emitted and retracted in all directions. After a couple of hours, by a process the reverse of swallow-

ing, the worm was slowly ejected, quite dead, and much changed in appearance, a large proportion of its contents having apparently been sucked out. For some hours subsequently the Rhizopod displayed remarkable activity, the pseudopodial filaments stretching out on all sides until they eventually entirely filled the field of a 3 in. objective, the centre being occupied by the creature itself. The movement of the sarcode to and fro both in its body and in every ramification of the pseudopodia was much increased in rapidity by the unwonted feast, and it was not until six or seven hours had elapsed that the rapidity and force of the streams were sensibly diminished, after which the animal gradually assumed an almost dormant condition.

In conclusion, I may just relate that in transferring this specimen from the bottle to the cell, I had some difficulty in detaching it from the pencil point, and in doing so accidentally separated two small portions of the naked external protoplasm. These detached pieces began as soon as they were settled in the cell to put out pseudopodia in all directions, until at last no central mass was left in either. Each had become an interlacing network of pseudopodia. The parent *Lieberkühnia* at the same time began to put out its pseudopodia, which ultimately reached those extended from the detached portions, and the moment actual contact took place they coalesced, the fragments again becoming part of the original body—thus repeating what had been previously observed and followed a stage further in *Shepheardella*.

Chester, *March*, 1880.

DEVELOPMENT *of the* KIDNEY *in its relation to the* WOLFFIAN
BODY *in the* CHICK. By ADAM SEDGWICK, B.A., Scholar
of Trinity College, Cambridge; Demonstrator in the
Morphological Laboratory.   (With   Plates   XVII,
XVIII.)

THIS paper contains an account of observations on the
development of the excretory system of the chick, made with
a view of elucidating the relation which the kidney bears to
the Wolffian body.

The Wolffian body in the embryo chick attains to a very
great development, but almost completely atrophies in the
adult, a small part only persisting in the male as part of the
testicular apparatus.

In the embryos of lower Vertebrates, viz. most of the
Icthyopsida, there is present, similarly, an organ called the
Wolffian body, which, however, much more completely
persists in the adult, functioning in part as kidney and in
part as semen carrier.

The separation into an urinary part and into a sexual part
is much more complete in some forms than in others. In
the Elasmobranchii, for instance, the posterior part of the
embryonic Wolffian body gives rise in the adult to a well-
developed gland, the kidney, while the anterior part attains
a far less development; in fact, more or less retrogrades in
the adult ; but in the male a part of it enters into connection
with the testis.

In the Amniota the case is different. In them an embryonic
organ, called the Wolffian body, does not function at all in
the adult as an excretory organ; it almost completely atro-
phies from its embryonic perfection, only a small part per-
sisting in the adult male as the epidydimis. The organ
which functions as kidney in the adult arises at a relatively
late stage, and is not apparently, as in Elasmobranchs, a
modified part of the hind end of the embryonic Wolffian
body. What, then, is the kidney of the Amniota? Is it an
organ which has arisen *de novo* in the Amniota, or can it, by
a more accurate study of its development, be traced into
relation with the embryonic excretory system? In other
words, can any evidence be obtained by the study of develop-
ment proving that the kidney of the chick phylogenetically
has been modified from part of the same primitive organ as
that from which the Wolffian body developed, as is the case
in the Icthyopsida ?

To obtain an answer to these questions I have been obliged to make a close study of the earliest stages in the development of the kidney and Wolffian body. The results obtained with regard to the latter are so different from those obtained by the latest observers, that I have recorded them in full in the following account.

Peculiarities in the early development of the Avian Woffian body necessitated an examination of the early development of the Wolffian tubules in other Vertebrates. This examination I was enabled to make in the case of Elasmobranchii owing to the great kindness of Mr. Balfour, who placed at my disposal the whole of his Elasmobranch sections. The result of this examination was to convince me that the account given of the earliest stages in the development of the Elasmobranch Wolffian body is in some respects erroneous.

Before proceeding to an account of the observations made upon these heads it will be well to give a short historical account of the progress of our knowledge on this subject, *i. e.* the development of the Wolffian body and kidney.

The later views as to the homologies of the parts of the excretory system found in the different members of the Vertebrate group dates from the work of Balfour[1] and Semper[2] on the embryology of Elasmobranchs.

The independent discoveries of these two investigators gave an impulse to the study of the development of the organs in question in other animals, and as a result it has gradually become clearer as the embryology of more animals became known that a great similarity in the development of these organs characterised the Vertebrata.

The earlier observers, Remak[3] and Rathke,[4] maintained that the tubules of the Wolffian body developed independently of the Wolffian duct in a blastema of mesoblast cells adjoining the inner side of the duct.

Waldeyer, in his well-known work,[5] asserted, from his observations, that the tubules of the Wolffian body developed as outgrowths from the duct, and that the Malpighian bodies arose independently in the adjoining mesoblast. The views of other observers, before 1874, were identical with one or the other of these.

[1] 'Monograph on the Development of Elasmobranch Fishes.'
[2] 'Urogenitalsystem der Plagiostomen Arbeiten,' vol. ii.
[3] 'Entwickelung der Wirbelthiere,' &c.
[4] 'Entwickelungsgeschichte der Wirbelthiere,' Leipzig, 1861.
[5] 'Eierstock und Ei,' 1870.

Since 1874 the work of Götte[1] and Spengel[2] on Amphibia, Kölliker[3] on Aves, Braun[4] on Lacertilia, and Fürbringer on Teleostei, Amphibia, and Aves[5] has shewn that the excretory system of all these animals is developed on a type seen in its simplest form in Elasmobranchs.

Kölliker first discovered in Aves structures composed of strings of cells connected with the Wolffian duct and peritoneal epithelium, and placed just ventral and internal to the former. These he compared to the early segmental tubes described in Elasmobranchs. From the similarity of these structures to those seen in Elasmobranchs and from his own observations he was led to assert for them a development similar to that described for Elasmobranchs, viz. from segmental involutions of the body-cavity epithelium.

In this he was followed by Fürbringer,[6] except in a small detail, the latter observer denying that these cell strings had any lumen opening into the body-cavity.

So far as I know, no ideas as to the morphological meaning of the Amniote kidney was held before 1874.

Conflicting statements were then put forward by different observers with regard to the actual embryonic development. Remak[7] and Kölliker[8] maintained that the whole of the epithelium of the kidney tubules, including that of the collecting and secreting tubules and the Malpighian bodies, was derived from a simple outgrowth from the ureter.

The condensed mesoblast tissue which lies near the ureter and its offshoots, in their opinion, only gives rise to the connective and vascular elements of the kidney.

Kölliker has expressed this view in the second edition of his great work on the development of Vertebrates. Löwe[9] has also recently arrived at the same conclusion from his observations on Mammals.

[1] 'Entwickelungsgesch. d. Unke.'

[2] "Das Urogen. system d. Amphibien," 'Arb. a. d. Zool. Inst.' Wurzburg, Bd. 3, 1876.

[3] 'Ent. gesch. d. Menschen u. d. höheren Thiere.'

[4] "Das Urogen-system d. d. Einheimischen Reptilien,' Semper's 'Arbeiten,' Bd. 4.

[5] "Zur vergleichenden Anat. u. Entwickelungsgeschichte d. Excretionsorgane der Vertebraten," 'Morphol. Jahrbuch,' Bd. 4. The reader is referred to this admirable essay for the literature, and a complete account of our knowledge of the excretory organs of Vertebrates.

Loc. cit.

' Entwickelung der Wirbelthiere.'

[8] Loc. cit.

[9] 'Centralblatt für die Med. Wissenschaften,' Oct., 1879.

Kupffer,[1] Bornhaupt,[2] and Braun,[3] on the other hand, assert that the secretory tubules and Malpighian bodies are formed independently of the ureter in the condensed mesoblast tissue mentioned above, the outgrowths from the ureter merely giving rise to the collecting tubules.

I shall return, when I have described the kidney development in the chick, to a consideration and discussion of the various hypotheses which have been held concerning the Amniote kidney.

Development of Wolffian body.—The ages of the younger embryos from which the sections figured in the accompanying plates (XVII and XVIII) were taken are indicated by the number of protovertebræ. In the older embryos this was not possible. In most cases the place in the body, from which a section figured was taken, is indicated by the number of the segment[4] in which it occurred, counting the first segment behind the auditory involutions as the first.

These determinations have been made with some care by mounting all the sections in order, and then by observing the protovertebræ, arranging them into groups corresponding to each protovertebra, beginning the process always in front.

The observations here recorded do not extend to any part of the Wolffian body in front of the fourteenth segment, nor to the development of the Wolffian duct. I have made some observations on both these parts, but they are not yet sufficiently complete to enable me to understand certain remarkable appearances in their development. The Wolffian body, like most other organs, develops first of all in front and then gradually backwards, so that supposing the development behind were the same as in front, the process might be shewn by a series of sections from a single chick of the proper age. But this is not the case. In the chick the development of the Wolffian tubules behind is very different to that in front. This fact has apparently been overlooked by the most recent observers.

The development of the Wolffian body in the duck is much more completely similar throughout than in the chick, and reference will be at first made to figs. 2—5, taken from a duck embryo with thirty-one or thirty-two protovertebræ, in the following description.

[1] 'Arch. f. Mic. Anat.,' Bd. 1.

[2] 'Untersuchungen über d. Entwick. des Urogen. systems beim Hühnchen,' Diss. Inaug., Riga, 1867.

[3] Loc. cit.

[4] The term segment is used as equivalent to protovertebra, muscle plate.

The tubules of the Wolffian body do not develop from serial involutions of the peritoneal epithelium, but from the cells of the intermediate cell mass. The intermediate cell mass is so familiar to all students of Avian embryology that
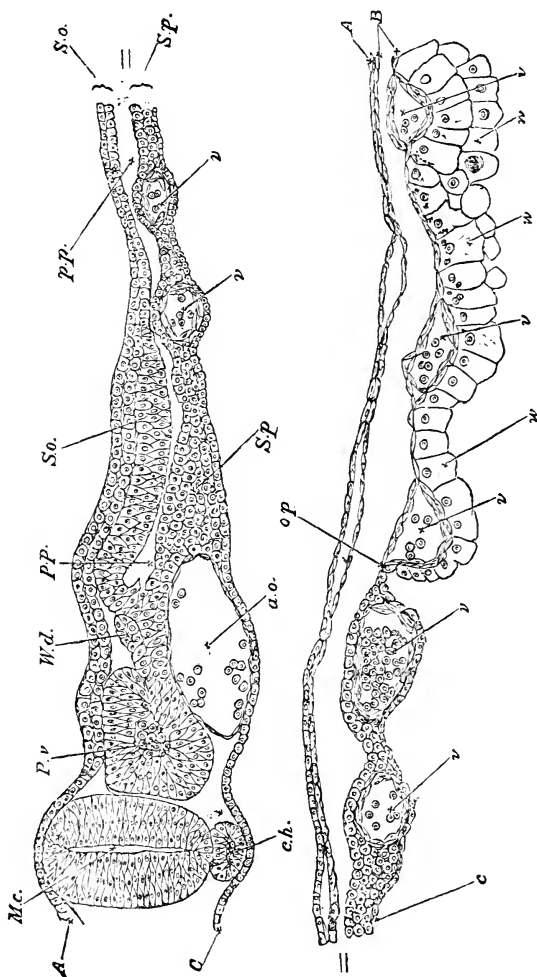
FIG. 1.[1]—Section from a chick of the second day, showing the intermediate cell-mass. *M. c.* Medullary canal. *P. v.* Protovertebra. *W. d.* Wolffian duct. *p. p.* Body-cavity. *c. h.* Notochord. *a. o.* aorta.

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[1] For the woodcuts, figs. 1 and 2, I have to thank Mr. Balfour. Fig. 1 is from the 'Elements of Embryology.'

it is hardly necessary to say anything about it. It is a mass of cells[1] (woodcut, fig. 1), stretching between the proto-vertebra (*P. v.*) and the dorsal inner angle of the body-cavity (*p.p.*). It is on its first formation continuous with the peritoneal epithelium. Its relation to the protovertebra is obscure, and I have been unable, so far, to make it out satisfactorily. There is one point, however, to be borne in mind concerning this intermediate cell mass; it is con-tinuous, *i. e.* is not divided by the lines of segmentation into areas corresponding to each protovertebra.

Very soon after the intermediate cell mass is established it undergoes a change. It becomes at some points more dis-tinctly continuous with the peritoneal epithelium, at others less so. And finally this culminates in a clear continuity, as seen in fig. 2, and a marked discontinuity, as seen in fig. 3. In fig. 2 we have what practically amounts to a continuation of the body-cavity into the intermediate cell mass (*i c m.*); in fig. 3, on the other hand, the intermediate cell mass is distinctly disconnected with the peritoneal epithelium, and lies as a mass of cells between it and the protovertebra. Al-though these figures are not taken from contiguous sections, fig. 2 being taken from the thirtieth segment, and fig. 3 from the twenty-ninth, yet for all the important details fig. 3 represents exactly a section next or next but one to fig. 2. The intermediate cell mass is then now present as a cord of cells continuous from segment to segment, and continuous at intervals with the peritoneal epithelium. Fig. 4 repre-sents the two conditions of the intermediate cell mass, as seen in a single section taken from the twenty-sixth seg-ment.

At the next stage of development the intermediate cell mass entirely breaks away from the peritoneal epithelium, and lies as a cellular blastema just internal to the Wolffian duct. It may be called the Wolffian blastema.

The Wolffian blastema almost directly breaks up into the structures constituting the first rudiments of the Wolffian tubules (fig. 5).

The development of the Wolffian blastema in the chick needs further description.

In the anterior region of the Wolffian body, as far back as the nineteenth or twentieth segments, the above description of the conversion of the intermediate cell mass into the

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[1] The term intermediate cell mass in this account is only used to in-dicate the cell mass connecting a *protovertebra* with the peritoneal epi-thelium, and never refers to the cell mass occupying the same position before segmentation has given rise to protovertebræ.

Wolffian blastema applies (*vide* fig. 1); but behind this region the development is different.

Here, even before the segments are formed, it is found that the cell mass, which on segmentation gives rise to the intermediate cell mass, is distinctly separate from the thick epithelium of the body-cavity, but attached to the cell mass, which will give rise to the protovertebra (fig. 6, taken from a chick with twenty-six protovertebræ behind the last protovertebra).

And this separation is apparently retained through later development. The cell mass (*i c m'*, fig. 6), in the next stage (fig. 7, taken from twenty-ninth segment of a chick with twenty-nine protovertebræ) is obviously the Wolffian blastema which, in a still later stage (fig. 10, from the twenty-ninth segment of a chick with thirty-four), gives rise to the commencing Wolffian tubule.

The same fact may be seen by comparing figs. 8 and 9, fig. 8 being taken from the twenty-fourth segment of a chick with twenty-six, and fig. 9 from the twenty-fourth segment of a chick with twenty-nine.

It will be observed, by inspection of figs. 6, 7, 8, that the peritoneal epithelium which adjoins the Wolffian blastema is thick, as it is elsewhere; while later (figs. 9, 10) it is in the same spot thin, as it is, or will be, in most other parts of the body-cavity.

No doubt this thick epithelium does, in the process of becoming thin, bud off cells, which travel inwards, and some of which may help to form the definite Wolffian blastema (figs. 9, 10). But this process takes place everywhere. In fact, at an early stage of development almost all the mesoblast cells are represented by the thick lining of the body-cavity ; and it is by a process of growth inwards of cells from this that most of the connective tissue, &c., of the wall of the body and gut is derived ; and, therefore, from the analogy of the fate of the cells growing out from the body-cavity wall in other places, we might fairly assume that those growing out from that particular part adjoining the intermediate cell mass or Wolffian blastema give rise, not to the Wolffian tubules, but to the connective tissue and blood-vessels of the Wolffian body. This is rendered highly probable from a consideration of some observations I have made on the development of the segmental tubes in Elasmobranchs, which point to the conclusion that the epithelial lining is derived from the cells of the intermediate cell mass. However that may be, of one fact there can be no doubt, viz. the cells from which the Wolffian tubules develop are not derived from

serial ingrowths of the body-cavity epithelium, for this thinning of the peritoneal epithelium, adjoining the region where they will appear, is continuous, *i. e.* cells must grow in along a line extending the whole length of that part of the body-cavity which the Wolffian body adjoins.

The development of the Wolffian blastema, so described, is continued as far back as the opening of the Wolffian duct into the cloaca, which occurs in the thirty-fourth segment. In fig. 12 it may be seen in the thirty-second segment (*k b*).

The Wolffian blastema of the chick then develops in two slightly different ways.

In the anterior part, about as far back as the twentieth (fig. 1) segment, that process of development which has been described at length in the case of the duck (in which animal it is apparently the only method of development) is passed through in the chick.

Posteriorly from the twentieth segment the intermediate cell mass has never any connection with the peritoneal epithelium, and gives rise to the Wolffian blastema quite independently of the peritoneal epithelium. This latter process is clearly an abbreviation of that which takes place throughout in the duck and in the anterior part of the chick.

I have mentioned the twentieth segment as *about* the limit between the two. I cannot fix the exact limit.

It has been stated above that in the case of the duck and the anterior part of the chick (figs. 1, 2, 4) the intermediate cell mass becomes, at certain points, very markedly continuous with the peritoneal epithelium, and appears to enclose a prolongation of the body-cavity (fig. 1 and fig. 2). Such connections are undoubtedly rudiments of the nephrostomata seen in other Vertebrates. They do not occur segmentally, being situated as often as not between the protovertebræ.

Rudiments of these rudimentary nephrostomata occur in the posterior part of the chick's Wolffian body ; that is, although a fairly sharp line can always be drawn between the Wolffian blastema and the peritoneal epithelium, yet the cells of the latter, at certain points, arrange themselves just as they do in front, where the line of the body-cavity is continued into the intermediate cell mass. These latter rudiments are very obscure, and I have been unable to make any satisfactory determination of their number. They may be due merely to an accidental arrangement of the cells, which might occur in consequence of the bend in the peritoneal epithelium at this point. Whether the rudimentary nephrostomata in the anterior part of the chick's kidney

(fig. 1[1]) are continued as small channels in the intermediate cell mass or Wolffian blastema, and remain, enlarging when the Wolffian tubules are formed later, I have been unable to ascertain. Neither have I been able to satisfy myself as to another interesting point, viz. Do those rudimentary nephrostomata correspond to the Wolffian tubules subsequently developed? In the chick's twentieth segment never more than three, at the most, nephrostomata can be made out, yet there are four or five primary Wolffian tubules later.

Has this increase been caused by the development of more tubules than there were nephrosomata, *i. e.* by intercalation, or has it been caused by a change in the relation of the parts to one another, due either to an elongation of the proto-vertebra or to the travelling forward of the tubules as they are developed behind?

I shall return to the consideration of this point in a future paper.

The mode in which the Wolffian blastema behind the twentieth[2] segment breaks up into tubules, so far as I have been able to ascertain it, is the following:—A number of vesicles, either oval or circular, lined by columnar cells, and lying just internal to the Wolffian duct, make their appearance (fig. 5). In longitudinal sections it may be seen that these vesicles closely adjoin one another antero-posteriorly. They are developed from those cells of the Wolffian blastema immediately adjoining the inner border of the Wolffian duct. By a study of transverse sections it appears that each vesicle is continuous ventrally with that part of the Wolffian blastema which has not undergone conversion into the walls of the vesicle, and which lies just internal to the vesicle. The cells of this part of the Wolffian blastema very soon arrange themselves round what appears as a continuation of the original vesicle (fig. 11). From the inner and dorsal wall of the last-formed structure a glomerulus is ultimately developed. The whole structure grows enormously, and gives rise to the Malpighian body and complicated coils of the later Wolffian tubule. At about the stage of development represented in fig. 11 the tubules acquire an opening into the Wolffian duct.

The question as to whether or no there are outgrowths from the Wolffian duct to meet the independently developed

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[1] *Vide* also fig. 125, of Kölliker's 'Entwick. gesch. der Menschen u. der. h. Thiere.'

[2] I reserve an account of the development of the tubules in front of the twentieth segment, as my observations on this point are not yet sufficiently complete to enable me to speak with certainty.

Wolffian tubules is not easy to answer. I leave it open now, but hope to be in a position to give a definite answer soon. Now I will merely state that there are appearances in my sections which incline me to the opinion that there are outgrowths from the Wolffian duct which, in the case of the primary Wolffian tubule, are solid, but hollow in the case of the secondary and tertiary tubules.

The above description of the development of the primary Wolffian tubules differs from the most recent account of Kölliker[1] and Fürbringer.[2] I have stated above the views which these distinguished observers hold as to the development. They have described perfectly correctly one stage in the development of the anterior part of the Wolffian body. I have often seen the appearances given by Kölliker in fig. 125 of his work, and have given myself a similar representation (fig. 1). But if I understand them correctly they have given an erroneous account of the earlier development of these structures. Fürbringer says of them (p. 67): " Sie finden sich in reihenweiser Anordnung als solide Urnierenstränge die von dem parietalen Peritoneum ventral und medial vom Wolff'schen Gange ausgehen. . . . Sehr bald lösen sich diese Urnierenstränge von dem parietalen Peritoneum ab und liegen nun als rundliche solide Zellenmassen retroperitoneal neben dem Wolff'schen Gange, ein Stadium das die Beobachtungen der mersten Autoren deckt." It is easy to see how Fürbringer has been misled. He has seen in transverse sections in a fairly young chick the S-shaped strings of cells (solide Urnierenstränge) in connection with the peritoneal epithelium. He has also seen in an older chick the Wolffian blastema of the posterior region of the Wolffian body (rundliche solide Zellenmassen). Both these observations I can entirely confirm. But apparently he has not examined the condition of these structures at an earlier stage, assuming that they originate as solid outgrowths of the peritoneal epithelium. This assumption my observations prove to be unwarranted.

The older observers (see above) were quite correct in their statements of the origin of the Wolffian tubules, as structures developed in the intermediate cell mass, independently of the Wolffian duct, and later acquiring an opening into it.

Waldeyer's[3] statement that the Malpighian body thus develops, the rest of the Wolffian tubule developing as outgrowths from the Wolffian duct, is in my opinion erroneous. For if there be an outgrowth from the Wolffian duct it does not give rise to the whole tubule, exclusive of the

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[1] Loc. cit.   [2] 'Morph. Jahrbuch,' Bd. 4.   [3] Loc. cit.

Malpighian body, the structure developed independently in the intermediate cell mass certainly giving rise to more than the Malpighian body.

I now pass to the development of the secondary tubules, &c. Fürbringer[1] derives them also from peritoneal ingrowths. He has not, however, given, so far as I know, any figures showing this. I have examined this point with some care, but have quite failed to discover any traces of these secondary ingrowths.

The secondary tubules appear to me to arise from a small mass of cells. They occupy, at a slightly earlier stage, the position of *wt.*[2] in fig. 11. In this figure the vesicular rudiment of a secondary tubule has appeared in this mass of cells. The tertiary and quaternary, &c., tubules appear to arise successively at a slightly later stage from similar small masses of cells, which are always placed just dorsal to the last-formed tubule. The later development of these secondary, tertiary, &c., Wolffian tubules is very similar to that of the primary.

The time of development of the primary tubules relatively to that of the secondary, &c., tubules varies in different parts. Anteriorly the primary tubule is much more developed (in fact, has acquired an opening into the Wolffian duct, not figured in fig. 11), before the first trace of the secondary tubule arises (fig. 11) ; while posteriorly a secondary and tertiary tubule have appeared almost before the primary tubule has lost its vesicular structure (fig. 13).

The development of the secondary, &c., Wolffian tubules in the chick appears to be very much abbreviated.

Whatever may have been their development in phylogeny, no light is thrown upon it by their ontogeny. Nor even can a comparison be made between their development in the chick, and that in other forms in which it is possible to suppose the development is less abbreviated. In Elasmobranchs the secondary tubules, as Balfour[2] has shewn, develop in connection with the Malpighian bodies of the primary tubules, as outgrowths from them, which eventually open into the collecting tubules of the segment in front. Neither Balfour nor, as far as I know, any other observer, have elucidated the development of the tertiary, &c., tubules in Elasmobranchs.

In the Salamander Fürbringer[3] has shewn that they develop as they do in the chick from cell masses closely

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[1] Loc. cit.
[2] Balfour, 'Elasmobranch Fishes.'
[3] Loc. cit.

adjoining the primary tubules, and from an inspection of his figures it is evident that these cell masses are situated close to the Malpighian body of the primary tubule. In the chick I have sought in vain for some clear sign in the development of these cells which would enable a comparison to be instituted with Elasmobranch development.

In the chick the cells of the Wolffian blastema do not all seem to be used in the formation of the primary tubule. Those, that are not, seem to collect at a special point, *i. e.* just dorsal to that part of the primary tubule which will become eventually the Malpighian body. The cells of the primary tubule are especially thick at this point, and perhaps they give rise to some of the cells for the secondary tubule (fig. 11). Even if this is the case—and we may look upon the secondary tubule as an outgrowth from the primary tubule—it is impossible to say where the secondary tubule so formed opens into the Wolffian duct.

This brings me to another difference between the dorsal tubules of the chick and those of Elasmobranchs. In the latter they all open into the collecting part of a primary Wolffian tubule. In the former they all open independently into the Wolffian duct, or it may be into an outgrowth from it, but separately from the primary tubule. This latter point I am, as above stated, obliged to leave open at present. The number of primary tubules present in one segment seem to be fairly constant, five or six to each segment, throughout the Wolffian body, except quite in front, where there seem to be fewer. All segments, from the twentieth to the thirtieth inclusive, contain five or six primary tubules. In front of the twentieth segment they seem gradually to decrease. In the first segment in which a fully developed tubule appears there seems only to be one, the number increasing rapidly to the twentieth.

The dorsal tubules appear in greater number behind than in front. In the twenty-eighth segment I have counted as many as four, but more are possibly developed later. They correspond in number to the primary tubules, *i. e.* if there are five primary tubules in the twenty-eighth segment, there are twenty secondary tubules (five sets of four). The most anterior segment in which a secondary tubule appears is the twenty-first or twenty-second; I have not been able, however, to localise it exactly.

Development of kidney.—The development of the Wolffian blastema from the intermediate cell mass has been described as far back as the thirty-fourth segment; *i. e.* to the opening of the Wolffian duct into the cloaca; it is never seen

behind this point.   But it does not all undergo the above development into Wolffian tubules.   It breaks up into Wolffian tubules as far back as the thirtieth segment.   Behind this point, *i. e.* from the thirty-first to the thirty-fourth segments inclusively, the Wolffian blastema undergoes quite a different fate.   It remains for some time almost quite passive and ultimately gives rise to the epithelium of the permanent kidneys.   In consequence of this I have called that part of the Wolffian blastema between the thirty-first and thirty-fourth segments the kidney blastema ; and in future shall refer to it by that name (figs. 12, 15, 16, 17, *kb*).   It is important to notice that this kidney blastema develops in an exactly similar manner to the Wolffian blastema.

It is not until well into the fourth day, when the ureter has appeared, that it is possible to draw the line between the two.

Fig. 12 is taken from the thirty-second segment of a chick with thirty-four protovertebræ; it shows a blastema of cells lying just internal to the Wolffian duct.   Fig. 10 is taken from the twenty-ninth segment of the same chick.   It shows the hindermost trace of a Wolffian tubule I could find at this stage.   In all the sections between figs. 10 and 12 there is present, just as in fig. 12, a blastema of cells lying just internal to the Wolffian duct.

In a slightly older embryo the hindermost trace of a Wolffian tubule would be in the thirtieth segment.   In still older embryos secondary tubules would have appeared in the thirtieth segment, but no trace of a primary tubule in the thirty-first, and so on in later stages, Wolffian tubules never appearing in the thirty-first segment.   In the embryo, from which figs. 13 to 17 were taken, the ureter had not appeared. In examining a series of sections from the posterior part of this embryo, some of which are figured (figs. 13 to 17), the following points are noticeable, illustrating what has just been stated.

A primary and secondary tubule are present in fig. 14, and it is almost the last section in which any trace of a Wolffian tubule can be seen (the two above tubules are cut in the next two sections).   The tubules adjacent anteriorly to these are three in number (fig. 13), consisting of primary, secondary, and tertiary.   Supposing the Wolffian body were going on developing in the region behind that from which fig. 14 was taken, we ought at the least to find at this stage primary tubules in that region, for the formation of primary and secondary tubules is always separated by an interval of time. But no such primary tubules are seen.   Behind fig. 14 (figs.

15 to 17) a blastema of cells is still present, precisely similar in its appearance and position with regard to the Wolffian duct to the Wolffian blastema seen anteriorly at earlier stages, and to the blastema seen in the same region at later stages (fig. 12). And this can be traced back to the opening of the Wolffian duct into the cloaca (fig. 17).

To this blastema of cells I have given the name kidney blastema; it is at this stage perfectly continuous anteriorly with the hinder Wolffian tubules, the junction between the two lying in one of the two sections intervening between figs. 14 and 15. I ought rather to say the line of future separation, for so far they have been always continuous, having developed so. The continuity between the kidney blastema (*kb*) and the hinder part of the Wolffian body may be seen in fig. 21, which is taken from a chick of nearly the same age as that from which figs. 13—17 were.

In this figure the section has passed through the hind end of the Wolffian duct and through the kidney blastema, and has just shaved the hinder end of the Wolffian body, in consequence of which the hinder Wolffian tubules are only indistinctly visible.

The next change to notice is caused by the appearance of the ureter. It arises as a growth forward from the dorsal border of the enlarged end of the Wolffian duct. This has been generally recognised since Kupffer's[1] account. The dilatation of the hind end of the Wolffian duct occurs in a very slightly later embryo than that from which fig. 21 was taken.

The kidney blastema is now found not ventrally close to the body-cavity, but lies dorsal to its former position, just internal to the dorsal extremity of the dilated Wolffian duct (fig. 20). From this dilatation there grows forward a duct, the ureter (fig. 19), on the inner side of which lies the kidney blastema.

The ureter, at this stage, has not a very great extent, and is only seen for a few more sections; in fig. 18, still behind the Wolffian body, the ureter is not visible; but the kidney blastema occupies a dorsal position, as it did in the posterior section in which the ureter was present (fig. 19).

In tracing it forward it gradually descends and becomes continuous with the hind end of the Wolffian body.

In yet older embryos, in which the ureter is more developed and overlaps the hind end of the Wolffian body, the kidney blastema has quite broken off from the Wolffian body, and invests the anterior end of the ureter, so that in a series of transverse sections through a chick at this age we should see

[1] ' Arch. f. M. Anat.,' Bd. 2.

posteriorly, in the same section with the now complicated Wolffian body, a dorsal mass of cells. Gradually travelling backwards a duct would appear cut across lying in the mass of cells; further back still we should see no Wolffian body, but merely a duct with a mass of cells lying just internal to it, placed well dorsal to the Wolffian duct. This mass of cells is the kidney blastema; and the duct is the ureter.

Such would be seen in a chick at the end of the fifth day.

On the sixth day the ureter grows in length, the kidney blastema accompanying it, and enveloping its anterior extremity.

The ureter now dilates at intervals, and the kidney blastema especially collects round these dilatations. From the latter, the number of which I have not determined, the kidney tubules grow out. In a chick of the seventh day the tubules are just beginning to grow out from these dilatations. The two posterior tubules are, however, far more advanced than the anterior.

The ureter is now a small duct lying just dorsal to the Wolffian body; except at its anterior extremity, where it is rather more dorsal, and is completely surrounded by the kidney blastema.

Almost immediately in front of the hind end of the ureter a tubule is given off, which runs dorsalwards and outwards. The kidney blastema no longer adjoins the ureter, but is disposed round the branches of this tubule. The ureter is continued forwards through a considerable number of sections, giving off no tubules, and unaccompanied by the kidney blastema. It now becomes continuous with a tubule, which has already been seen in many sections surrounded by kidney blastema, and which, though not so much branched as the most posterior tubule above mentioned, is more developed than any tubule met with in front.

The ureter continues as a small duct lying just dorsal to the Wolffian body.

In this embryo (seventh day), travelling forwards, several dilations could be made out. The appearance presented by such a dilatation in transverse section and its position with regard to the Wolffian body, may be gathered from an inspection of fig. 22.

The lateral walls of the dorsal part of the dilated ureter are closely applied, the lumen being very indistinct.

Around the dorsal part of the dilatation the kidney blastema is present.

In the next section, or in the next section but one, either

backwards or forwards, the dorsal dilatation would be no longer visible, but occupying the position of the dorsal part of the dilatation, would be seen a tubule surrounded by the cells of the kidney blastema (fig. 23).

In the next section to this the walls of the tubule become indistinctly marked off from the kidney blastema (fig. 24). Some of the large columnar cells of the kidney tubule become branched, the processes being continuous with the processes of the branched cells of the kidney blastema. In fact, every stage of cell shape between a columnar lining cell of the tubule and a branched cell of the blastema is visible.

The lumen of the tubule is no longer distinct, it not being possible to say what is an intercellular space and what the lumen of the tubule.

In the next section no trace of a tubule is visible, its place being occupied by the cells of the blastema.

Fig. 23 is taken from a section next but one to fig. 22. Nine such dorsal dilatations of the ureter, with commencing tubules, growing from them, could be made out in the embryo under consideration.

In front of the most anterior the tubules open directly into the ureter which in this region has become more dorsal with regard to the Wolffian body. Tubules in this region, moreover, are given off from the ventral side of the ureter, corresponding almost exactly to those given off from the dorsal side. Four pairs could be made out; after which the ureter ended closely surrounded on all sides by dense kidney blastema.

The next stage, which I have closely examined, was in an eight-day chick. The kidney had reached a great complication of structure. Malpighian bodies had, however, not distinctly appeared. The tubules were still surrounded by kidney blastema which was especially conspicuous at their growing ends. The appearance of the latter, which was exactly similar in all essential details to the growing points of the tubule last described, is represented in fig. 24.

Before considering the bearing of the above facts upon the questions asked at the outset, I will recapitulate the more important points in the development of the Avian kidney and Wolffian body.

1. The cells which give rise to the Wolffian and kidney tubules do not develop as involutions of the peritoneal epithelium, but from a blastema of cells derived from the intermediate cell mass.

2. The blastema of the kidney is at first perfectly continuous with that of the Wolffian body, and cannot be distinguished from it.

3. Wolffian tubules do not appear in any part of the blastema behind the thirtieth segment. Primary, secondary, and tertiary, &c., tubules are developed in that part of it placed in the thirtieth and anterior segments as far as the twenty-first or twenty-second, and primary tubules in yet anterior segments.

4. The blastema in the thirty-first to thirty-fourth segments, on the appearance of the ureter, moves dorsalwards from the Wolffian duct, breaking away from the hindermost Wolffian tubules and enters into close relation with the ureter.

5. This part of the blastema—the kidney blastema—especially collects round swellings on the ureter, from which kidney tubules grow out.

6. These kidney tubules burrow into the kidney blastema, their growing points being continuous with the cells of the blastema.

Five years ago Balfour [1] and Semper [2] independently put forward the hypothesis that the kidney of the Amniota holds the same relation to the embryonic Wolffian body as does the adult kidney in Elasmobranchs.

Balfour wrote then :[3] " The last feature in the anatomy of the Selachians which requires notice is the division of the kidney into two portions, an anterior and posterior. The anatomical similarity between this arrangement and that of higher Vertebrates (birds, &c.) is very striking. The anterior one precisely corresponds, anatomically, to the *Wolffian body,* and the posterior to the true permanent *kidney* of higher Vertebrates ; and when we find that in the Selachians the duct for the anterior serves also for the semen, as does the duct of higher Vertebrates, this similarity seems almost to amount to identity."

The development of the kidney of the bird has never been fully worked out, so that this hypothesis, arrived at from a consideration of the facts of comparative anatomy and Elasmobranch embryology, has hitherto not been tested by the facts of Avian embryology. The observations described above were undertaken with a view of testing this hypothesis, and, in my opinion, it has fully stood the test.

The development of the kidney in the chick points most decidedly to the conclusion that it is merely the posterior part of the Wolffian body—or, perhaps, it would be better to say, of a primitive organ, the anterior part of which is now

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[1] " Urogenital Organs of Vertebrates," 'Journal of Anatomy and Physiology,' vol. x.
[2] Loc. cit.
[3] P. 27.

seen as the Wolffian body—the whole of this primitive organ in Elasmobranch embryos being termed Wolffian body.

The most important fact in favour of Balfour's hypothesis is the primitive continuity in early stages in the bird of the cells from which both Wolffian body and kidney arise.

The differences in later development cannot be looked upon as a serious difficulty when we remember the immense differences which many undoubtedly homologous organs show in their embryonic development in various animals.

It has been stated (see above, p. 148) by many students of Avian embryology that the kidney tubules develop as outgrowths from the ureter, and that the cells of the kidney blastema merely give rise to the vascular elements of the glomerulus. This view, whether considered à priori, or with reference to the facts of development, cannot for a moment be maintained.

If Balfour's hypothesis as to the relation of the kidney to the Wolffian body be accepted, and I do not see how it can be rejected, assuming the truth of the facts of development recorded in this account, it would require very strong proof indeed to establish the fact that the cells of the kidney blastema give rise merely to the vascular elements of the glomerulus, and take no part in the formation of the secretory epithelium of the kidney tubules, such as is taken in the formation of tubules of a very similar organ by cells developed in a precisely similar way. Such proof is not forthcoming, and would be very hard to give.

Considering the very late development of the posterior part of the Wolffian body (kidney) in the chick with reference to that of the anterior part, it surely cannot be a matter of surprise if the development has been modified, the walls of the tubule arising from the cells of the blastema; the lumen, however, not as in the anterior part, first appearing as an independent cavity, which opens later into the duct, but being from the first continuous with the lumen of the ureter.

Fürbringer's suggestion, that the Amniote kidney is derived from dorsal tubules of the Wolffian body, is based mainly on the fact that it lies dorsal to the Wolffian body, and an observation of Braun's for Lizards. Braun has stated for these animals that the kidney blastema develops from irregular ingrowths of the peritoneal epithelium, at a period when the secondary dorsal tubules of the Wolffian body are developing.

With regard to the first point it is to be noted that the

dorsal position of the kidney is clearly a secondary change, appearing only late in development, and due obviously to the great size the kidney attains. Moreover, according to Fürbringer's view, one would expect to find some kind of continuity between the developing kidney and dorsal part of the Wolffian body ; but no trace of any such connection can ever be seen.

Finally, in view of the facts of development here recorded for the chick, and of those about to be mentioned for Elasmobranchs, Braun's observations on the development of the kidney blastema of Lizards from peritoneal ingrowths cannot be accepted without further evidence. The irregularly scattered cells lying between the Wolffian duct and the peritoneal epithelium, which Braun has figured, are by no means proof of ingrowth of cells from the peritoneal epithelium. Such an irregular arrangement of cells can be seen anywhere adjoining the body-cavity epithelium.

Kölliker's view that the kidney of the Amniota is an organ *sui generis,* which was not present in any form in the excretory system of the common ancestor of Icthyopsida and Amniota, needs in my opinion no refutation ; for if true it can only be established by proving all other hypotheses concerning the kidney to be untenable.

Development of segmental tubes in Elasmobranchii.—I should hardly have been bold enough to publish these observations on the development of the chick's Wolffian body, opposed as they are to statements supported by great authority, had I not had the opportunity of examining the early development of the parts in question in Elasmobranchs.

I was thus enabled to confirm suspicions which I had entertained since examining the development of the Wolffian body of birds, as to the correctness of the description of the earliest stages in these fishes. It is well known that the Wolffian tubules of Elasmobranchii are derived from the segmentally-arranged segmental tubes. These latter were said to arise by an invagination, at first solid but subsequently becoming hollow, of the peritoneal epithelium just internal to the segmental duct into the cells of the intermediate cell mass. The intermediate cell mass was said to be produced by the coming together of the splanchnic and somatic layers of that part of the body cavity, which at an earlier period existed connecting the general ventral body cavity with the dorsal continuations of it in the muscle plates.

On examining specimens of young Elasmobranch (Scyllium, Pristiurus, Torpedo) embryos, I found that the passage connecting the general body-cavity with that in the

muscle-plates persisted later than had been described. Its connection with the ventral dilatation of the muscle-plate cavity is carried ventralwards as far as the outer dorsal corner of the segmental duct; so that it appears as a canal opening into the body-cavity just internal to the segmental duct, and thence curling round its dorsal wall to open into the muscle-plate cavity. The ventral outer wall of this passage is formed of large columnar cells, the inner and dorsal wall of much flatter cells (woodcut, fig. 2), as seen in transverse sections.
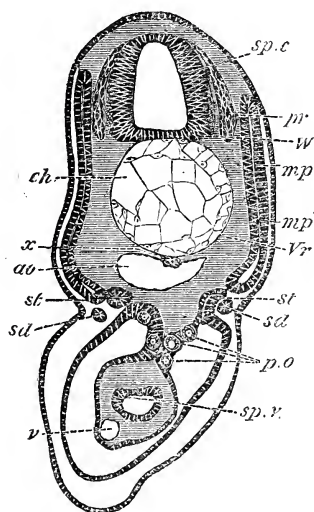
FIG. 2.[1]—Transverse section through a young embryo of Scytlium. *mp.* Muscle-plate. *st.* Segmental tube. *sd.* Segmental duct. *ch.* Notochord. *sp. v.* Alimentary canal.

At the next stage of development the passage becomes quite separated from the muscle-plate cavity, and now lies as a blind tube, opening into the body-cavity internal to the segmental duct, its blind outer end being applied to the ventral dilatation of the muscle-plate body-cavity (woodcut, fig. 2). This blind tube is the commencement of a segmental tube.

I have traced it through a series of older embryos to the fully-formed segmental tube. The adjoining woodcut (fig. 2)

[1] Fig. 2 is from Mr. Balfour's forthcoming treatise on 'Comparative Embryology.' It is copied from a figure in his 'Monograph on the Development of Elasmobranch Fishes.'

represents a section from a Scytlium embryo, in which the segmental tube has just broken away from the muscle plate ; in a slightly younger embryo, or perhaps in posterior sections of the same embryo, the cavity of the segmental tube (*st*) would communicate with the ventral dilatation of the muscle plate (*mp*), at the point where they are in contact in the figure.

This mode of origin of the segmental tubes of Elasmobranchii renders the origin of the same structures in the chick less extraordinary than it at first sight seemed.

I refrain now from the discussions and perhaps hypotheses which this observation on the development of the Elasmobranch segmental tubes suggests. On one point, however, there can be little doubt, viz. that segmental involutions of the peritoneal epithelium which have been described in the Teleostei, Amphibia, Ganoids, Mammals, will have to be given up. At any rate the current statement on this point cannot be accepted without further proof.

I have made observations on the Teleostei and Sturgeon (Mr. Balfour having kindly put his specimens at my disposal) which tend to show, not, however, with certainty, that in the embryos of these forms at any rate there are no serial involutions of the body-cavity epithelium to form the segmental tubes. The method of development in these forms appears to me to be very much modified, if we continue to regard the development in Elasmobranchs as primitive.

The Wolffian body in all those animals whose ova have a relatively small food yolk seems to be retarded in development ; while the head kidney, the relation of which to the rest of the excretory system is not understood, attains an early development and functions as the larval excretory organ. Very possibly a clue to the explanation of the retardation in the development of the Wolffian body and of the morphological meaning of the head kidney, in Teleostei, Amphibia, &c., may be obtained by a consideration of this coincidence and others hitherto apparently overlooked.

I hope in another paper to discuss these questions and some others which have been passed over here, and to describe the development of the anterior part of the Wolffian body in the chick.

In conclusion, I wish to acknowledge the great obligations I am under to Mr. Balfour. Not only have I to thank him for his kindness in placing all his preparations and specimens at my disposal, but, for what has been far more valuable, the help and encouragement he has given me through the whole course of this investigation.

NOTES *on the* DEVELOPMENT *of the* ARANEINA.   By F. M.
BALFOUR, M.A., F.R.S., Fellow of Trinity College,
Cambridge.   With Plates XIX, XX, and XXI.

THE following observations do not profess to contain a
complete history of the development even of a single species
of spider.   They are the result of investigations carried on
at intervals during rather more than two years, on the ova
of *Angelena labyrinthica ;* and I should not have published
them now, if I had any hope of being able to complete them
before the appearance of the work I am in the course of
publishing on Comparative Embryology.   It appeared to me,
however, desirable to publish in full such parts of my
observations as are completed before the appearance of my
treatise, since the account of the development of the Aran-
eina is mainly founded upon them.

My investigations on the germinal layers and organs have
been chiefly conducted by means of sections.   To prepare
the embryos for sections, I employed the valuable method
first made known by Bobretsky.   I hardened the embryos
in bichromate of potash, after placing them for a short time
in nearly boiling water.   They were stained as a whole
with hematoxylin after the removal of the membranes, and
embedded for cutting in coagulated albumen.

The number of investigators who have studied the de-
velopment of spiders is inconsiderable.   A list of them is
given at the end of the paper.

The earliest writer on the subject is Herold (No. 4) ; he
was followed after a very considerable interval of time by
Claparède (No. 3), whose memoir is illustrated by a series
of beautiful plates, and contains a very satisfactory account
of the external features of development.

Balbiani (No. 1) has gone with some detail into the
history of the early stages ; and Ludwig (No. 5) has pub-
lished some very important observations on the development
of the blastoderm.   Finally, Barrois (No. 2) has quite re-
cently taken up the study of the group, and has added some
valuable observations on the development of the germinal
layers.

In addition to these papers on the true spiders, important
investigations have been published by Metschnikoff on
other groups of the Arachnida, notably the scorpion.
Metschnikoff's observations on the formation of the ger-

minal layers and organs accord in most points with my own.

The development of the Araneina may be divided into four periods: (1) the segmentation; (2) the period from the close of the segmentation up to the period when the segments commence to be formed; (3) the period from the commencing formation of the segments to the development of the full number of limbs; (4) the subsequent stages up to the attainment of the adult form.

In my earliest stage the segmentation was already completed, and the embryo was formed of a single layer of large flattened cells enveloping a central mass of polygonal yolk-segments.

Each yolk-segment is formed of a number of large clear somewhat oval yolk-spherules. In hardened specimens the yolk-spherules become polygonal, and in ova treated with hot water prior to preservation are not unfrequently broken up. Amongst the yolk-segments are placed a fair number, of nucleated bodies of a very characteristic appearance. Each of them is formed of (1) a large, often angular, nucleus, filled with deeply staining bodies (nucleoli ?). (2) Of a layer of protoplasm surrounding the nucleus, prolonged into a protoplasmic reticulum. The exact relation of these nucleated bodies to the yolk-segments is not very easy to make out, but the general tendency of my observations is to show (1) that each nucleated body belongs to a yolk-sphere, and (2) that it is generally placed not at the centre, but to one side of a yolk-sphere. If the above conclusions are correct each complete yolk-segment is a cell, and each such cell consists of a normal nucleus, protoplasm, and yolk-spherules. There is a special layer of protoplasm surrounding the nucleus, while the remainder of the protoplasm consists of a reticulum holding together the yolk-spherules. Yolk-cells of this character are seen in Pl. XX and XXI, figs. 10—21.

The nuclei of the yolk-cells are probably derived by division from the nuclei of the segmentation rosettes (*vide* Ludwig, No. 5), and it is probable that they take their origin at the time when the superficial layer of protoplasm separates from the yolk-columns below to form the blastoderm.

The protoplasm of the yolk-cells undergoes rapid division, as is shewn by the fact that there are often two nucleated bodies close together, and sometimes two nuclei in a single mass of protoplasm (fig. 10). It is probable that in some cases the yolk-spheres divide at the same time as the protoplasm belonging to them; the division of the nucleated

bodies is, however, in the main destined to give rise to fresh cells which enter the blastoderm.

I have not elucidated to my complete satisfaction the next stage or two in the development of the embryo; and have not succeeded in completely reconciling the results of my own observations with those of Claparède and Balbiani.   In order to show exactly where my difficulties lie it is necessary briefly to state the results arrived at by the above authors.

According to Claparède the first differentiation in Pholcus consists in the accumulation of the cells over a small area to form a protuberance, which he calls the *primitive cumulus.* Owing to its smaller specific gravity the part of the ovum with the cumulus always turns upwards, like the blasto-dermic pole of a fowl's egg.

After a short time the cumulus elongates itself on one side, and becomes connected by a streak with a white patch, which appears on the surface of the egg, about 90° from the cumulus.   This patch gradually enlarges, and soon covers the whole surface of the ovum except the region where the cumulus is placed. It becomes the ventral plate or germinal streak of the embryo, its extremity adjoining the cumulus is the anal extremity, and its opposite extremity the cephalic one.   The cumulus itself is placed in a depression on the dorsal surface of the ovum.   Claparède compares the cumulus to the dorsal organ of many Crustacea.

Balbiani (No. 1) describes the primitive cumulus in *Tegenaria domestica, Epeira diadema,* and *Agelena labyrin-thica,* as originating as a protuberance at the centre of the ventral surface, surrounded by a specialised portion of the blastoderm (p. 57), which I will call the ventral plate.   In *Tegenaria domestica* he finds that it encloses the so-called yolk-nucleus, p. 62.   By an unequal growth of the ventral plate the primitive cumulus comes to be placed at the cephalic pole of the ventral plate.   The cumulus now becomes less prominent, and in a few cases disappears.   In the next stage the central part of the ventral plate becomes very prominent and forms the procephalic lobe, close to the anterior border of which is usually placed the primitive cumulus (p. 67).   The space between the cumulus and the procephalic lobe grows larger, so that the latter gradually travels towards the dorsal surface and finally vanishes. Behind the procephalic lobe the first traces of the segments make their appearance, as three transverse bands, before a distinct anal lobe becomes apparent.

The points which require to be cleared up are, (1) what is the nature of the primitive cumulus? (2) where is it

situated in relation to the embryo? Before attempting to answer these questions I will shortly describe the development, so far as I have made it out, for the stages during which the cumulus is visible.

The first change that I find in the embryo (when examined after it has been hardened)[1] is the appearance of a small, whitish spot, which is at first very indistinct. A section through such an ovum (Pl. XX, fig. 10) shows that the cells of about one half of the ovum have become more columnar than those of the other half, and that there is a point (*pr. c.*) near one end of the thickened half where the cells are more columnar, and about two layers or so deep. It appears to me probable that this point is the whitish spot visible in the hardened ovum. In a somewhat later stage (Pl. XIX, fig. 1) the whitish spot becomes more conspicuous (*pc*), and appears as a distinct prominence, which is, without doubt, the primitive cumulus, and from it there proceeds on one side a whitish streak. The prominence, as noticed by Claparède and Balbiani, is situated on the flatter side of the ovum. Sections at this stage show the same features as the previous stage, except that (1) the cells throughout are smaller, (2) those of the thickened hemisphere of the ovum more columnar, and (3) cumulus is formed of several rows of cells, though not divided into distinct layers. In the next stage the appearances from the surface are rather more obscure, and in some of my best specimens a coagulum, derived from the fluid surrounding the ovum, covers the most important part of the blastoderm. In Pl. XIX, fig. 2, I have attempted to represent, as truly as I could, the appearances presented by the ovum. There is a well-marked whitish side of the ovum, near one end of which is a prominence (*pc*), which must, no doubt, be identified with the cumulus of the earlier stages. Towards the opposite end, or perhaps rather nearer the centre of the white side of the ovum, is an imperfectly marked triangular white area. There can be no doubt that the line connecting the cumulus with the triangular area is the future long axis of the embryo, and the white area is, without doubt, the procephalic lobe of Balbiani.

A section of the ovum at this stage is represented in Pl. XX, fig. 11. It is not quite certain in what direction the section is taken, but I think it probable it is somewhat oblique to the long axis. However this may be, the section

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[1] I was unfortunately too much engaged, at the time when the eggs were collected, to study them in the fresh condition; a fact which has added not a little to my difficulties in elucidating the obscure points in the early stages.

shows that the whitish hemisphere of the blastoderm is formed of columnar cells, for the most part two or so layers deep, but that there is, not very far from the middle line, a wedge-shaped internal thickening of the blastoderm where the cells are several rows deep. With what part visible in surface view this thickened portion corresponds is not clear. To my mind it most probably corresponds to the larger white patch, in which case I have not got a section through the terminal prominence. In the other sections of the same embryo the wedge-shaped thickening was not so marked, but it, nevertheless, extended through all the sections. It appears to me probable that it constitutes a longitudinal thickened ridge of the blastoderm. In any case, it is clear that the white hemisphere of the blastoderm is a thickened portion of the blastoderm, and that the thickening is in part due to the cells being more columnar, and, in part, to their being more than one row deep, *though they have not become divided into two distinct germinal layers*. It is further clear that the increase in the number of cells in the thickened part of the blastoderm is, *in the main, a result of the multiplication of the original single row of cells*, while a careful examination of my sections proves that it is also partly due to cells, derived from the yolk, having been added to the blastoderm.

In the following stage which I have obtained (which cannot be very much older than the previous stage, because my specimens of it come from the same batch of eggs), a distinct and fairly circumscribed thickening forming the ventral surface of the embryo has become established. Though its component parts are somewhat indistinct, it appears to consist of a procephalic lobe, a less prominent caudal lobe, and an intermediate portion divided into about three segments; but its constituents cannot be clearly identified with the structures visible in the previous stage. I am inclined, however, to identify the anterior thickened area of the previous stage with the procephalic lobe, and a slight protuberance of the caudal portion (visible from the surface) with the primitive cumulus. I have, however, failed to meet with any trace of the cumulus in my sections.

To this stage, which forms the first of the second period of the larval history, I shall return, but it is necessary now to go back to the observations of Claparède and Balbiani.

There can, in the first place, be but little doubt that what I have called the primitive cumulus in my description is the structure so named by Claparède and Balbiani.

It is clear that Balbiani and Claparède have both failed to appreciate the importance of the organ, which my observations show to be the part of the ventral thickening of the blastoderm where two rows of cells are first established, and therefore the point where the first traces of the future mesoblast becomes visible.

Though Claparède and Balbiani differ somewhat as to the position of the organ, they both make it last longer than I do : I feel certainly inclined to doubt whether Claparède is right in considering a body he figures after six segments are present, to be the same as the dorsal organ of the embryo before the formation of any segments, especially as all the stages between the two appear to have escaped him. In Agelena there is undoubtedly no organ in the position he gives when six segments are found.

Balbiani's observations accord fairly with my own up to the stage represented in fig. 2. Beyond this stage my own observations are not satisfactory, but I must state that I feel doubtful whether Balbiani is correct in his description of the gradual separation of the procephalic lobe and the cumulus, and the passage of the latter to the dorsal surface, and think it possible that he may have made a mistake as to which side of the procephalic lobe, in relation to the parts of the embryo, the cumulus is placed.

Although there appear to be grounds for doubting whether either Balbiani and Claparède are correct in the position they assign to the cumulus, my observations scarcely warrant me in being very definite in my statements on this head, but, as already mentioned, I am inclined to place the organ near the posterior end (and therefore, as will be afterwards shown, in a somewhat dorsal situation) of the ventral embryonic thickening.

In my earliest stage of the third period there is present, as has already been stated, a procephalic lobe, and an indistinct and not very prominent caudal portion, and about three segments between the two. The definition of the parts of the blastoderm at this stage is still very imperfect, but from subsequent stages it appears to me probable that the first of the three segments is that of the first pair of ambulatory limbs, and that the segments of the chelicerae and pedipalpi are formed later than those of the first three ambulatory appendages.

Balbiani believes that the segment of chelicerae is formed later than that of the six succeeding segments. He further concludes, from the fact that this segment is cut off from the procephalic portion in front, that it is really part of the

procephalic lobe. I cannot accept the validity of this argument; though I am glad to find myself in, at any rate, partial harmony with the distinguished French embryologist as to the facts. Balbiani denies for this stage the existence of a caudal lobe. There is certainly, as is very well shown in my longitudinal sections, a thickening of the blastoderm in the caudal region, though it is not so prominent in surface views as the procephalic lobe.

A transverse section through an embryo at this stage (Pl. XX, fig. 12) shows that there is a ventral plate of somewhat columnar cells more than one row deep, and a dorsal portion of the blastoderm formed of a single row of flattened cells. Every section at this stage shows that the inner layer of cells of the ventral plate is receiving accessions of cells from the yolk, which has not to any appreciable extent altered its constitution. A large cell, passing from the yolk to the blastoderm, is shown in fig. 12 at *y. c.*

The cells of the ventral plate are now divided into two distinct layers. The outer of these is the *epiblast,* the inner the *mesoblast.* The cells of both layers are quite continuous across the median line, and exhibit no trace of a bilateral arrangement.

This stage is an interesting one on account of the striking similarity which (apart from the amnion) exists between a section through the blastoderm of a spider and that of an insect immediately after the formation of the mesoblast. The reader should compare Kowalevsky's (' Mem. Acad. Petersbourg,' vol. xvi, 1871) fig. 26, Pl. IX with my fig. 12. The existence of a continuous ventral plate of mesoblast has been noticed by Barrois (p. 532), who states that the two mesoblastic bands originate from the longitudinal division of a primitive single band.

In a slightly later stage (Pl. XIX, fig. 3 *a* and 3 *b*) six distinct segments are interpellated between the procephalic and the caudal lobes. The two foremost, *ch* and *pd* (especially the first), of these are far less distinct than the remainder, and the first segment is very indistinctly separated from the procephalic lobe. From the indistinctness of the first two somites, I conclude that they are later formations than the four succeeding ones. The caudal and procephalic lobes are very similar in appearance, but the procephalic lobe is slightly the wider of the two. There is a slight protuberance on the caudal lobe, which is possibly the remnant of the cumulus. The superficial appearance of segmentation is produced by a series of transverse valleys, separating raised intermediate portions which form the segments. The

ventral thickening of the embryo now occupies rather more
than half the circumference of the ovum.

Transverse sections show that considerable changes have
been effected in the constitution of the blastoderm. In the
previous stage, the ventral plate was formed of an uniform
external layer of epiblast, and a continuous internal layer
of mesoblast. The mesoblast has now become divided along
the whole length of the embryo, except, perhaps, the proce-
phalic lobes, into two lateral bands which are not continuous
across the middle line (Pl. XX, fig. 13 me). It has, more-
over, become a much more definite layer, closely attached
to the epiblast. Between each mesoblastic band and the
adjoining yolk there are placed a few scattered cells, which
in a somewhat later stage become the splanchnic mesoblast.
These cells are derived from the yolk-cells; and almost
every section contains examples of such cells in the act of
joining the mesoblast..

The epiblast of the ventral plate has not, to any great
extent, altered in constitution. It is, perhaps, a shade
thinner in the median line than it is laterally. The division
of the mesoblast plate into two bands, together, perhaps,
with the slight reduction of the epiblast in the median
ventral line, gives rise at this stage to an imperfectly
marked median groove.

The dorsal epiblast is still formed of a single layer of flat
cells. In the neighbourhood of this layer the yolk nuclei
are especially concentrated. The yolk itself remains as
before.

The segments continue to increase regularly, each fresh
segment being added in the usual way between the last
formed segment and the unsegmented caudal lobe. At the
stage when about nine or ten segments have become estab-
lished, the first rudiments of appendages become visible.
At this period (Pl. XIX, fig. 4) there is a distinct median
ventral groove, extending through the whole length of the
embryo, which becomes, however, considerably shallower
behind. The procephalic region is distinctly bilobed. The
first segment (that of the cheliceræ) is better marked off
. from it than in the previous stage, but is without a trace
of an appendage, and exhibits therefore, in respect to the
development of its appendages, the same retardation that
characterised its first appearance. The next five segments,
viz. those of the pedipalpi and four ambulatory appendages,
present a very well-marked swelling at each extremity.
These swellings are the earliest traces of the appendages.
Of the three succeeding segments, only the first is well

differentiated. The caudal lobe, though less broad than the procephalic lobe, is still a widish structure. The most important internal changes concern the mesoblast, which is now imperfectly though distinctly divided into somites, corresponding with segments visible externally. Each meso-blastic somite is formed of a distinct somatic layer closely attached to the epiblast, and a thinner and less well-marked splanchnic layer. In the appendage-bearing segments the somatic layer is continued up into the appendages.

The epiblast is distinctly thinner in the median line than at the two sides.

The next stage figured (Pl. XIX, figs. 5 and 6) is an important one, as it is characterised by the establishment of the full number of appendages. The whole length of the ventral plate has greatly increased, so that it embraces nearly the circumference of the ovum, and there is left uncovered but a very small arc between the two extremities of the plate (Pl. XIX, fig. 6; Pl. XX, fig. 15). This arc is the future dorsal portion of the embryo, which lags in its development immensely behind the ventral portion.

There is a very distinctly bilobed procephalic region (*pr. l*) well separated from the segment with the cheliceræ (*ch*). It is marked by a shallow groove opening behind into a circular depression (*st.*)—the earliest rudiment of the stomodæum. The six segments behind the procephalic lobes are the six largest, and each of them bears two prominent appendages. They constitute the six appendage-bearing segments of the adult. The four future ambulatory appendages are equal in size: they are slightly larger than the pedipalpi, and these again than the cheliceræ. Behind the six somites with prominent appendages there are four well-marked somites, each with a small protuberance. These four protuberances are provisional appendages. They have been found in many other genera of Araneina (Claparède, Barrois). The segments behind these are rudimentary and difficult to count, but there are, at any rate, five, and at a slightly later stage probably six, including the anal lobe. These fresh segments have been formed by the continued segmentation of the anal lobe, which has greatly altered its shape in the process. The ventral groove of the earlier stage is still continued along the whole length of the ventral plate.

By the close of this stage the full number of post-cephalic segments has become established. They are best seen in the longitudinal section (Pl. XX, fig. 15). There are six anterior appendage-bearing segments, followed by four with rudimentary appendages (not seen in this figure), and six without

appendages behind.   There are, therefore, sixteen in all.
This number accords with the result arrived at by Barrois,
but is higher by two than that given by Claparède.

The germinal layers (*vide* Pl. XX, fig. 14) have by this
stage undergone a further development.   The mesoblastic
somites are more fully developed.   The general relations of
these somites is shown in longitudinal section in Pl. XX,
fig. 15, and in transverse section in Pl. XX, fig. 14.   In the
tail, where they are simplest (shown on the upper side in fig.
14), each mesoblastic somite is formed of a somatic layer of
more or less cubical cells attached to the epiblast, and a
splanchnic layer of flattened cells. Between the two is placed a
completely circumscribed cavity, which constitutes part of the
embryonic body cavity. Between the yolk and the splanchnic
layer are placed a few scattered cells, which form the latest
derivatives of the yolk-cells, and are to be reckoned as part of
the splanchnic mesoblast.   The mesoblastic somites do not
extend outwards beyond the edge of the ventral plate, and
the corresponding mesoblastic somites of the two sides do
not nearly meet in the middle line.   In the limb-bearing
somites the mesoblast has the same general characters as in
the posterior somites, but the *somatic* layer is prolonged as
a hollow papilliform process into the limb, so that each limb
has an axial cavity continuous with the section of the body
cavity of its somite.   The description given by Metschnikoff
of the formation of the mesoblastic somites in the scorpion,
and their continuation into the limbs, closely corresponds
with the history of these parts in spiders.   In the region of
each procephalic lobe the mesoblast is present as a continuous
layer underneath the epiblast, but in the earlier part of the
stage, at any rate, is not formed of two distinct layers with
a cavity between them.

The epiblast at this stage has also undergone important
changes.   Along the median ventral groove it has become
very thin.   On each side of this groove it exhibits in each
appendage-bearing somite a well-marked thickening, which
gives in surface views the appearance of a slightly raised
area (Pl. XIX, fig. 5), between each appendage and the
median line.   These thickenings are the first rudiments of
the ventral nerve ganglia.   The ventral nerve cord at this
stage is formed of two ridge-like thickenings of the epiblast,
widely separated in the median line, each of which is con-
stituted of a series of raised divisions—the ganglia—united by
shorter, less prominent divisions (fig. 14, *vg*).   The nerve
cords are formed from before backwards, and are not at this
stage found in the hinder segments.   *There is a distinct*

*ganglionic thickening for the chelicerœ quite independent of
the procephalic lobes.*

In the procephalic lobes the epiblast is much thickened
and is formed of several rows of cells. The greater part o.
it is destined to give rise to the supra-œsophageal ganglia.

During the various changes which have been described the
blastoderm cells have been continually dividing, and, together
with their nuclei, have become considerably smaller than at
first. The yolk cells have in the meantime remained much
as before, and are, therefore, considerably larger than the
nuclei of the blastoderm cells. They are more numerous
than in the earlier stages, but are still surrounded by a
protoplasmic body, which is continued into a protoplasmic
reticulum. The yolk is still divided up into polygonal seg-
ments, but from sections it would appear that the nuclei are
more numerous than the segments, though I have failed to
arrive at quite definite conclusions on this point.

As development proceeds the appendages grow longer and
gradually bend inwards. They become very soon divided by
a series of ring-like constrictions which constitute the first
indications of the future joints (Pl. XIX, fig. 6). The full
number of joints are not at once reached, but in the ambu-
latory appendages five only appear at first to be formed.
There are four joints in the pedipalpi, while the cheliceræ do
not exhibit any signs of becoming jointed till somewhat later.
The primitive presence of only five joints in the ambulatory
appendages is interesting, as this number is permanent in
Insects and in Peripatus.

The next stage figured forms the last of the third
period (Pl. XIX, fig. 7 and 7 *a*). The ventral plate
is still rolled round the egg (fig. 7), and the end of the tail
and the procephalic lobes nearly meet dorsally, so that there
is but a very slight development of the dorsal region. There
are the same number of segments as before, and the chief
differences in appearance between the present and the
previous stage depend upon the fact (1) that the median
ventral integument between the nerve ganglia has become
wider, and at the same time thinner ; (2) that the limbs have
become much more developed ; (3) that the stomodæum is
definitely established ; (4) that the procephalic lobes have
undergone considerable development.

Of these features, the three last require a fuller descrip-
tion. The limbs of the two sides are directed towards each
other, and nearly meet in the ventral line. The cheliceræ
are two-jointed, and terminate in what appear like rudi-
mentary chelæ, a fact which perhaps indicates that the

spiders are descended from ancestors with chelate cheliceræ. The four embryonic post-ambulatory appendages are now at the height of their development.

The stomodæum (Pl. XIX, fig. 7, and Pl. XX, fig. 17, *st*) is a deepish pit between the two procephalic lobes, and distinctly in front of segment of the cheliceræ. It is bordered in front by a large, well-marked, bilobed upper lip, and behind by a smaller lower lip. The large upper lip is a temporary structure, to be compared, perhaps, with the gigantic upper lip of the embryo of Chelifer (cf. Metschinkoff). On each side of and behind the mouth two whitish masses are visible, which are the epiblastic thickenings which constitute the ganglia of the cheliceræ (Pl. XIX, fig. 7, *ch. g*).

The procephalic lobes (*pr. l*) now form two distinct masses, and each of them is marked by a semicircular groove, dividing them into a narrower anterior and a broader posterior division.

In the region of the trunk the general arrangement of the germinal layers has not altered to any great extent. The ventral ganglionic thickenings are now developed in all the segments in the abdominal as well as in the thoracic region. The individual thickenings themselves, though much more conspicuous than in the previous stage (Pl. XX, fig. 16, *v. c*), are still integral parts of the epiblast. They are more widely separated than before in the middle line. The mesoblastic somites retain their earlier constitution (Pl. XX, fig. 16). Beneath the procephalic lobes the mesoblast has, in most respects, a constitution similar to that of a mesoblastic somite in the trunk. It is formed of two bodies, one on each side, each composed of a splanchnic and somatic layer (Pl. XX, fig. 17, *sp.* and *so*), enclosing between them a section of the body-cavity. But the cephalic somites, unlike those of the trunk, are united by a median bridge of mesoblast, in which no division into two layers can be detected. This bridge assists in forming a thick investment of mesoblast round the stomodæum (*st*).

The existence of a section of the body cavity in the præoral region is a fact of some interest, especially when taken in connection with the discovery, by Kleinenberg, of a similar structure in the head of Lumbricus. The procephalic lobe represents the præoral lobe of Chætopod larvæ, but the prolongation of the body cavity into it does not, in my opinion, necessarily imply that it is equivalent to a post-oral segment.

The epiblast of the procephalic lobes is a thick layer several cells deep, but without any trace of a separation of the ganglionic portion from the epidermis.

The nuclei of the yolk have increased in number, but the yolk, in other respects, retains its earlier characters.

The next period in the development is that in which the body of the embryo gradually acquires the adult form. The most important event which takes place during this period is the development of the dorsal region of the embryo, which, up to its commencement, is practically non-existent. As a consequence of the development of the dorsal region, the embryo, which has hitherto had what may be called a dorsal flexure, gradually unrolls itself, and acquires a ventral flexure. This change in the flexure of the embryo is in appearance a rather complicated phenomenon, and has been somewhat differently described by the two naturalists who have studied it in recent times.

For Claparède the prime cause of the change of flexure is the translation dorsalwards of the limbs. He compares the dorsal region of the embryo to the arc of a circle, the two ends of which are united by a cord formed by the line of insertion of the limbs. He points out that if you bring the middle of the cord, so stretched between the two ends of the arc, nearer to the summit of the arc, you necessarily cause the two ends of the arch to approach each other, or, in other words, if the insertion of the limbs is drawn up dorsally, the head and tail must approach each other ventrally.

Barrois takes quite a different view to that of Claparède, which will perhaps be best understood if I quote a translation of his own words. He says: "At the period of the last stage of the embryonic band (the stage represented in Pl. XX, fig. 7, in the present paper) this latter completely encircles the egg, and its posterior extremity nearly approaches the cephalic region. Finally, the germinal bands, where they unite at the anal lobe (placed above on the dorsal surface), form between them a very acute angle. During the following stages one observes the anal segment separate further and further from the cephalic region, and approach nearer and nearer to the ventral region. This displacement of the anal segment determines, in its turn, a modification in the divergence of the anal bands ; the angle which they form at their junction tends to become more obtuse. The same processes continue regularly till the anal segment comes to occupy the opposite extremity to the cephalic region, a period at which the two germinal bands are placed in the same plane and the two sides of the obtuse angle end by meeting in a straight line. If we suppose a continuation of the same phenomenon it is clear that the

anal segment will come to occupy a position on the ventral surface, and the germinal bands to approach, but in the inverse way, so as to form an angle opposite to that which they formed at first. This condition ends the process by which the posterior extremity of the embryonic band, at first directed towards the dorsal side, comes to bend in towards the ventral region."

Neither of the above explanations is to my mind perfectly satisfactory. The whole phenomenon appears to me to be very simple, and to be caused by the elongation of the dorsal region, *i. e.* the region on the dorsal surface between the anal and procephalic lobes. Such an elongation necessarily separates the anal and procephalic lobes; but, since the ventral plate does not become shortened in the process, and the embryo cannot straighten itself on account of the egg-shell, it necessarily becomes flexed, and such flexure can only be what I have already called a ventral flexure. If there were but little food yolk this flexure would cause the whole embryo to be bent in, so as to have the ventral surface concave, but instead of this the flexure is confined at first to the two bands which form the ventral plate. These bands are bent in the natural way (Pl. XIX, fig. 8, в), but the yolk forms a projection, a kind of yolk sack as Barrois calls it, distending the thin integument between the two ventral bands. This yolk sack is shown in surface view in Pl. XIX, fig. 8, and in section in Pl. XXI, fig. 18. At a later period, when the yolk has become largely absorbed in the formation of various organs, the true nature of the ventral flexure becomes apparent, and the abdomen of the young Spider is found to be bent over so as to press against the ventral surface of the thorax (Pl. XIX, fig. 9). This flexure is shown in section in Pl. XXI, fig. 21.

At the earliest stage of this period of which I have examples, the dorsal region has somewhat increased, though not very much. The limbs have grown very considerably and *now cross in the middle line.*

The ventral ganglia, though not the supra-œsophageal, have become separated from the epiblast.

The yolk nuclei, each surrounded by protoplasm as before, are much more numerous.

In other respects there are no great changes in the internal features.

In my next stage, represented in Pl. XIX, figs. 8 *a*, and 8 *b*, a very considerable advance has become effected. In the first place the dorsal surface has increased in length to rather more than one half the circumference of the ovum.

The dorsal region has, however, not only increased in length, but also in definiteness, and a series of transverse markings (fig. 8 *a* and *b*), which are very conspicuous in the case of the four anterior abdominal segments (the segments with rudimentary appendages), have appeared, indicating the limits of segments dorsally.   The terga of the somites may, in fact, be said to have become formed.   The posterior terga (fig. 8 *a*) are very narrow compared to the anterior.

The caudal protuberance is more prominent than it was, and somewhat bilobed ; it is continued on each side into one of the bands, into which the ventral plate is divided. These bands, as is best seen in side view (fig. 8 *b*), have a ventral curvature, or, perhaps more correctly, are formed of two parts, which meet at a large angle open towards the ventral surface.   The posterior of these parts bears the four still very conspicuous provisional appendages, and the anterior the six pairs of thoracic appendages.   The four ambulatory appendages are now seven-jointed, as in the adult, but though longer than in the previous stage they do not any longer *cross or even meet in the middle line*, but are, on the contrary, separated by a very considerable interval.   This is due to the great distension by the yolk of the ventral part of the body, in the interval between the two parts of the original ventral plate.   The amount of this yolk may be gathered from the section (Pl. XXI, fig. 18). The pedipalpi carry a blade on their basal joint.   The chelicerae no longer appear to spring from an independent postoral segment.

There is a conspicuous lower lip, but the upper is less prominent than before.   Sections at this stage show that the internal changes have been nearly as considerable as the external.

The dorsal region is now formed of a (1) flattened layer of epiblast cells, and a (2) fairly thick layer of large and rather characteristic cells which any one who has studied sections of spider's embryos will recognise as derivatives of the yolk. These cells are not, therefore, derived from prolongations of the somatic and splanchnic layers of the already formed somites, but are new formations derived from the yolk.   They commenced to be formed at a much earlier period, and some of them are shown in the long section (Pl. XX, fig. 15). In the next stage these cells become differentiated into the somatic and splanchnic mesoblast layers of the dorsal region of the embryo.

In the dorsal region of the abdomen the heart has already become established.   So far as I have been able to make

out it is formed from a solid cord of the cells of the dorsal region. The peripheral layer of this cord gives rise to the walls of the heart, while the central cells become converted into the corpuscles of the blood.

The rudiment of the heart is in contact with the epiblast above, and there is no greater evidence of its being derived from the splanchnic than from the somatic mesoblast; it is, in fact, formed before the dorsal mesoblast has become differentiated into two layers.

In the abdomen three or four transverse septa, derived from the splanchnic mesoblast, grow a short way into the yolk. They become more conspicuous during the succeeding stage, and are spoken of in detail in the description of that stage. In the anterior part of the thorax a longitudinal and vertical septum is formed, which grows downwards from the median dorsal line, and divides the yolk in this region into two parts. In this septum there is formed at a later stage a vertical muscle attached to the suctorial part of the stomodæum.

The mesoblastic somites of the earlier stage are but little modified; and there are still prolongations of the body cavity into the limbs (Pl. XXI, fig. 18).

The lateral parts of the ventral nerve cords are now at their maximum of separation (Pl. XXI, fig. 18, $v. g$). Considerable differentiation has already set in in the constitution of the ganglia themselves, which are composed of an outer mass of ganglion cells enclosing a kernel of nerve fibres, which lie on the inner side and connect the successive ganglia. There are still distinct thoracic and abdominal ganglia for each segment, and there is also a pair of separate ganglion for the cheliceræ, which assists, however, in forming the œsophageal commissures.

The thickenings of the præoral lobe which form the supra-œsophageal ganglia are nearly though not quite separated from the epiblast. The semicircular grooves of the earlier stages are now deeper than before, and are well shown in sections nearly parallel to the outer anterior surface of the ganglion (Pl. XXI, fig. 19). The supra-œsophageal ganglia are still entirely formed of undifferentiated cells, and are without commissural tissue like that present in the ventral ganglia.

The stomodæum has considerably increased in length, and the proctodæum has become formed as a short, posteriorly directed involution of the epiblast. I have seen traces of what I believe to be two outgrowths from it, which form the Malpighian bodies.

The next stage constitutes (Pl. XIX, fig. 9) the last which

requires to be dealt with so far as the external features are concerned. The yolk has now mainly passed into the abdomen, and the constriction separating the thorax and abdomen has began to appear. The yolk sack has become absorbed, so that the two halves of the ventral plate in the thorax are no longer widely divaricated. The limbs have to a large extent acquired their permanent structure, and the rings of which they are formed in the earlier stages are now replaced by definite joints. A delicate cuticle has become formed, which is not figured in my sections. The four rudimentary appendages have disappeared, unless, which to seems me in the highest degree improbable, they remain as the spinning mammillæ, two pairs of which are now present. Behind is the anal lobe, which is much smaller and less conspicuous than in the previous stage. The spinnerets and anal lobe are shown as five papillæ in Pl. XIX, fig. 9. Dorsally the heart is now very conspicuous, and in front of the cheliceræ may be seen the supra-œsophageal ganglia.

The indifferent mesoblast has now to a great extent become converted into the permanent tissues. On the dorsal surface there was present in the last stage a great mass of unformed mesoblast cells. This mass of cells has now become divided into a somatic and splanchnic layer (Pl. XXI, fig. 22). It has, moreover, in the abdominal region at any rate, become divided up into somites. At the junction between the successive somites the splanchnic mesoblast on each side of the abdomen dips down into the yolk and forms a septum (Pl. XXI, fig. 22 s). The septa so formed, which were first described by Barrois, are not complete. The septa of the two sides do not, in the first place, quite meet along the median dorsal or ventral lines, and in the second place they only penetrate the yolk for a certain distance. Internally they usually end in a thickened border.

Along the line of insertion of each of these septa there is developed a considerable space between the somatic and splanchnic layers of mesoblast. The parts of the body cavity so established are transversely directed channels passing from the heart outwards. They probably constitute the venous spaces, and perhaps also contain the transverse aortic branches.

In the intervals between these venous spaces the somatic and splanchnic layers of mesoblast are in contact with each other.

I have not been able to work out satisfactorily the later stages of development of the septa, but I have found that they play an important part in the subsequent development

of the abdomen. In the first place they send off lateral off-shoots, which unite the various septa together, and divide up the cavity of the abdomen into a number of partially separated compartments. There appears, however, to be left a free axial space for the alimentary tract, the meso-blastic walls of which are, I believe, formed from the septa.

At the present stage the splanchnic mesoblast, apart from the septa, is a delicate membrane of flattened cells (fig. 22, *sp*). The somatic mesoblast is thicker, and is formed of scattered cells (*so*).

The somatic layer is in part converted, in the posterior region of the abdomen, into a delicate layer of longitudinal muscles, the fibres of which are not continuous for the whole length of the body, but are interrupted at the lines of junction of the successive segments. They are not present in the anterior part of the abdomen. The longitudinal direction of these fibres, and their division with myotomes, is interesting, since both these characters, which are preserved in Scorpions, are lost in the abdomen of the adult Spider.

The original mesoblastic somites have undergone quite as important changes as the dorsal mesoblast. In the abdominal region the somatic layer constitutes two powerful bands of longitudinal muscles, inserted anteriorly at the root of the fourth ambulatory appendage, and posteriorly at the spinning mammillæ. Between these two bands are placed the nervous bands. The relation of these parts are shown in the section in Pl. XXI, fig. 20 *d*, which cuts the abdomen horizontally and longitudinally. The mesoblastic bands are seen at *m.*, and the nervous bands within them at *ab. g.* In the thoracic region the part of the somatic layer in each limb is converted into muscles, which are continued into dorsal and ventral muscles in the thorax (*vide* fig. 20 *c*). There are, in addition to these, intrinsic transverse fibres on the ventral side of the thorax. Besides these muscles there are in the thorax, attached to the suctorial extremity of the stomodæum, three powerful muscles, which I believe to be derived from the somatic me-soblast. One of these passes vertically down from the dorsal surface, in the septum the commencement of which was described in the last stage. The two other muscles are lateral, one on each side (Pl. XX, fig. 20 *c*).

The heart has now, in most respects, reached its full development. It is formed of an outer muscular layer, within which is a doubly-contoured lining, containing nuclei at intervals, which is probably of the nature of an epithelioid lining (Pl. XXI, fig. 22 *ht*). In its lumen are numerous blood-corpuscles (not represented in my figure). **The heart**

lies in a space bound below by the splanchnic mesoblast, and to the sides by the somatic mesoblast. This space forms a kind of pericardium (fig. 22 *pc*), but dorsally the heart is in contact with the epiblast. The arterial trunks connected with it are fully established.

The nervous system has undergone very important changes.

In the abdominal region the ganglia of each side have fused together into a continuous cord (fig. 21 *ab.g.*). In fig. 20, in which the abdomen is cut horizontally and longitudinally, there are seen the two abdominal cords (*ab. g.*) united by two transverse commissures; and I believe that there are at this stage three or four transverse commissures at any rate, which remain as indications of the separate ganglia, from the coalescence of which the abdominal cords are formed. The two abdominal cords are parallel and in close contact.

In the thoracic region changes of not less importance have taken place. The ganglia are still distinct. The two cords formed of these ganglia are no longer widely separated in median line, but meet, in the usual way, in the ventral line. Transverse commissures have become established (fig. 20 *c*) between the ganglia of the two sides. There is as little trace at this, as at the previous stages, of an ingrowth of epiblast, to form a median portion of the central nervous system. Such a median structure has been described by Hatschek for Lepidoptera, and he states that it gives rise to the transverse commissures between the ganglia. My observations show that for the spider, at any rate, nothing of the kind is present.

As shown in the longitudinal section (Pl. XXI, fig. 21), the ganglion of the cheliceræ has now united with the supra-œsophageal ganglion. It forms, as is shown in fig. 20 *b* (*ch. g.*), a part of the œsophageal commissure, and there is no sub-œsophageal commissure uniting the ganglia of the cheliceræ, but the œsophageal ring is completed below by the ganglia of the pedipalpi (fig. 20 *c, pd. g.*).

The supra-œsophageal ganglia have become completely separated from the epiblast.

I have unfortunately not studied their constitution in the adult, so that I cannot satisfactorily identify the parts which can be made out at this stage.

I distinguish, however, the following regions:

(1) A central region containing the commissural part, and continuous below with the ganglia of the cheliceræ.

(2) A dorsal region formed of two hemispherical lobes.

(3) A ventral anterior region.

The central region contains in its interior the commissural portion, forming a punctiform, rounded mass in each ganglion. A transverse commissure connects the two (*vide* fig. 20 *b*).

The dorsal hemispherical lobes are derived from the part which, at the earlier stage, contained the semicircular grooves. When the supra-œsophageal ganglia become separated from the epidermis the cells lining these grooves become constricted off with them, and form part of these ganglia. Two cavities are thus formed in this part of the supra-œsophageal ganglia. These cavities become, for the most part, obliterated, but persist at the outer side of the hemispherical lobes (figs. 20 *a* and 21).

The ventral lobe of the brain is a large mass shown in long section in fig. 21. It lies immediately in front of and almost in contact with the ganglia of the cheliceræ.

The two hemispherical lobes agree in position with the fungiform body (pilzhutförmige Korpern), which has attracted so much the attention of anatomists, in the supra-œsophageal ganglia of Insects and Crustacea; but till the adult brain of Spiders has been more fully studied it is not possible to state whether the hemispherical lobes become fungiform bodies.

Hatschek[1] has described a special epiblastic invagination in the supra-œsophageal ganglion of Bombyx, which is probably identical with the semicircular groove of Spiders and Scorpions, but in the figure he gives the groove does not resemble that in the Arachnida. A similar groove is found in Peripatus, and there forms, as I have found, a large part of the supra-œsophageal ganglia. It is figured by Moseley, 'Phil. Trans.,' vol. 164, pl. lxxv, fig. 9.

The stomodæum is considerably larger than in the last stage, and is lined by a cuticle; it is a blind tube, the blind end of which is the suctorial pouch of the adult. To this pouch are attached the vertical dorsal, and two lateral muscles spoken of above.

The proctodæum (*pr.*) has also grown in length, and the two Malpighian vessels which grow out from its blind extremity (fig. 20 *e, mp. g.*) have become quite distinct. The part now formed is the rectum of the adult. The proctodæum is surrounded by a great mass of splanchnic mesoblast. The mesenteron has as yet hardly commenced to be developed. There is, however, a short tube close to the proctodæum (fig. 20 *e, mes*), which would seem to be the commencement of it. It

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[1] " Beiträge z. Entwick d. Lepidopteren," ' Jenaische Zeit.,' vol. xi, p. 124.

ends blindly on the side adjoining the rectum, but is open anteriorly towards the yolk, and there can be very little doubt that it owes its origin to cells derived from the yolk. On its outer surface is a layer of mesoblast.

From the condition of the mesenteron at this stage there can be but little doubt that it will be formed, not on the surface, *but in the interior of the yolk.* I failed to find any trace of an anterior part of the mesenteron adjoining the stomodæum. In the posterior part of the thorax (*vide* fig. 20 *d*), there is undoubtedly no trace of the alimentary tract.

The presence of this rudiment shows that Barrois is mistaken in supposing that the alimentary canal is formed entirely from the stomodæum and proctodæum, which are stated by him to grow towards each other, and to meet at the junction of the thorax and abdomen. My own impression is that the stomodæum and proctodæum have reached their full extension at the present stage, and that both the stomach in the thorax and the intestine in the abdomen are products of the mesenteron.

The yolk retains its earlier constitution, being divided into polygonal segments, formed of large yolk vesicles. The nuclei are more numerous than before. In the thorax the yolk is anteriorly divided into two lobes by the vertical septum, which contains the vertical muscle of the suctorial pouch. In the posterior part of the thorax it is undivided.

I have not yet been able clearly to make out the eventual fate of the yolk. At a subsequent stage, when the cavity of the abdomen is cut up into a series of compartments by the growth of the septa, described above, the yolk fills these compartments, and there is undoubtedly a proliferation of yolk cells round the walls of these compartments. It would not be unreasonable to conclude from this that the compartments were destined to form the hepatic cæca, each cæcum being enclosed in a layer of splanchnic mesoblast, and its hypoblastic wall being derived from the yolk cells. I think that this hypothesis is probably correct, but I have met with some facts which made me think it possible that the thickenings at the ends of the septa, visible in Pl. XXI, fig. 22, were the commencing hepatic cæca.

I must, in fact, admit that I have hitherto failed to work out satisfactorily the history of the mesenteron and its appendages. The firm cuticle of young spiders is an obstacle both in the way of making sections and of staining, which I have not yet overcome.

General Conclusions.

Without attempting to compare at length the development of the spiders with that of other Arthropoda, I propose to point out a few features in the development of spiders, which appear to show that the Arachnida are undoubtedly more closely related to the other Tracheata than to the Crustacea.

The whole history of the formation of the mesoblast is very similar to that in insects. The mesoblast in both groups is formed by a thickening of the median line of the ventral plate (germinal streak).

In insects there is usually formed a median groove, the walls of which become converted into a plate of mesoblast. In spiders there is no such groove, but a median keel-like thickening of the ventral plate (Pl. XX, fig. 11), is very probably an homologous structure. The unpaired plate of mesoblast formed in both insects and Arachnida is exactly similar, and beomes divided, in both groups, into two bands, one on each side of the middle line. Such differences as there are between Insects and Arachnida sink into insignificance compared with the immense differences in the origin of the mesoblast between either group, and that in the Isopoda, or, still more, the Malacostraca and most Crustacea. In most Crustacea we find that the mesoblast is budded off from the walls of an invagination, which gives rise to the mesenteron.

In both spiders and Myriopoda, and probably insects, the mesoblast is subsequently divided into somites, the lumen of which is continued into the limbs. In Crustacea mesoblastic somites have not usually been found, though they appear occasionally to occur, e. g. Mysis, but they are in no case similar to those in the Tracheata.

In the formation of the alimentary tract, again, the differences between the Crustacea and Tracheata are equally marked, and the Arachnida agree with the Tracheata. There is generally in Crustacea an invagination, which gives rise to the mesenteron. In Tracheata this never occurs. The proctodæum is usually formed in Crustacea before or, at any rate, not later than the stomodæum.[1] The reverse is true for the Tracheata. In Crustacea the proctodæum and stomodæum, especially the former, are very long, and usually give rise to the greater part of the alimentary tract, while the mesenteron is usually short.

[1] If Grobben's account of the development of Moina is correct this statement must be considered not to be universally true.

In the Tracheata the mesenteron is always considerable, and the proctodæum is always short. The derivation of the Malpighian bodies from the proctodæum is common to most Tracheata. Such organs are not found in the Crustacea.

With reference to other points in my investigations, the evidence which I have got that the cheliceræ are true postoral appendages supplied in the embryo from a distinct postoral ganglion, confirms the conclusions of most previous investigators, and shows that these appendages are equivalent to the mandibles, or possibly the first pair of maxilla of other Tracheata. The invagination, which I have found, of part of a groove of epiblast in the formation of the supraœsophageal ganglia is of interest, owing to the wide extension of a similar occurrence amongst the Tracheata.

The wide divarication of the ventral nerve cords in the embryo renders it easy to prove that there is no median invagination of epiblast between them, and supports Kleinenberg's observations on Lumbricus as to the absence of this invagination. I have further satisfied myself as to the absence of such an invagination in Peripatus. It is probable that Hatschek and other observers who have followed him are mistaken in affirming the existence of such an invagination in either the Chætopoda or the Arthropoda.

The observations recorded in this paper on the yolk cells and their derivations are, on the whole, in close harmony with the observations of Dohrn, Bobretsky, and Graber, on Insects. They show, however, that the first formed mesoblastic plate does not give rise to the whole of the mesoblast, but that during the whole of embryonic life the mesoblast continues to receive accessions of cells derived from the cells of the yolk.

Araneina.

1. *Balbiani*, "Mémoire sur le Développement des Araneides," 'Ann. Sci. Nat.,' series v, vol. xvii, 1873.

2. *J. Barrois*, "Recherches s. l. Développement des Araignées," 'Journal de l. Anat. e. de la Physiol.,' 1878.

3. *E. Claparède*, 'Recherches s. l'Évolution des Araignées,' Utrecht, 1860.

4. *Herold*, 'De Génératione Araniorum in Ovo,' Marburg, 1824.

5. *H. Ludwig*, "Ueb. d. Bildung des Blastoderm," bie d. Spinnea, 'Zeit. f. wiss. Zool.,' vol. xxvi, 1876.

A Contribution *to the* Biology *of* Bacteria.   By Dr. L.
     Waldstein, of the Pathological Institute, Heidelberg.[1]

THE experiments here recorded were not undertaken to meet
Dr. Bastian on his favourite field of research, nor does this
communication partake of the polemical nature of so many of
the recent publications on the same subject.   The wide import-
ance, quite irrespective of abiogenesis, which attaches itself
to Dr. Bastian's work,[2] is here recognised in so far as the method
employed by him is concerned.

Bacteria have been assigned a prominent position in pathology,
more especially since the theories of Pettenkofer and of Lister
have introduced them as causally connected with infection. They
are to-day certainly made responsible for more than can be well
proven with our present knowledge of the conditions of their
existence.   This can be gathered from the perusal of but a very
small portion of the voluminous literature, which has grouped
itself about the many theories and hypotheses as to their patho-
genic properties, based on experiments which, in many instances,
have given quite as frequently negative as positive results.

The study of the conditions under which Bacteria multiply,
as well as of those which prove to be unfavorable to their develop-
ment, is, clearly in view of these circumstances, of the highest
interest, and all researches in this direction ought to be examined
with great care, in an objective spirit.   Dr. Bastian, in the
course of his researches, has not strictly confined himself to these
questions alone.   He has also drawn conclusions which, as he
himself remarks, tend towards a complete revolution in the
current opinions held with respect to the rôle taken in pathology
by micro-organisms.

Dr. Bastian—citing the experiments of Gerhardt, which go to
prove that alcohol mixed with a little potash becomes converted
into vinegar and a brown resinous substance, whereas in its
natural state it can be exposed to the air indefinitely without
becoming acid—proposes to show that potash added to sterilised
and well guarded urine produces conditions favorable to the
spontaneous generation of micro-organisms. He adds the alkali
in a certain proportion, determined in each case by the acidity of
the urine.

[1] The author desires to refer the reader to his more extended memoir
on the same subject in Virchow's 'Archiv,' vol. 77, 1879.

[2] " On the Conditions favouring Fermentation, and the appearance of
Bacilli, Micrococci, and Torulæ in previously Boiled Fluids," by H. Charl-
ton Bastian. 'Journal of the Linnean Society (Zoology),' Oct. 24, 1877,
London.

I refer to the original for an account of the details of the experiment.

The result was invariably that the urine in the control experiments remains clear and apparently unaltered for an indefinite time; whilst where the potash had been allowed to act upon the sterilised fluid it became turbid, lighter in colour, and swarmed with organisms in from eighteen to twenty-six hours, on the average.

In repeating these experiments I have closely followed Dr. Bastian's directions, with the exception that the potash tubes were made by drawing out test tubes in the middle, charging them with the requisite amount of liquor potassæ, and then hermetically sealing them by melting, and thus separating the upper part of the tube. This procedure is much more convenient than the original method; and the sterilisation called for is ensured fully by keeping such tubes in boiling water during twenty minutes, as, says Dr. Bastian, in these experiments I soon found that the longer or shorter duration of the period of boiling of the liquor potassæ tubes did not appreciably influence the results.

Dr. W. Roberts and M. Pasteur could not obtain the same results, and it is furthermore not absolutely certain that all germs were kept out of the retorts when Dr. Bastian boiled the tubes in oil during a period of twenty hours. He himself mentions the difficulty of properly cleaning them then and, even if it be conceded that those contained within the tubes were destroyed, fresh germs might possibly adhere to the exterior, whereas the heat and the period of immersion of the urine retort in boiling water would be the only safeguard against a contamination with such germs.

If the views of all experimenters in regard to the degree of heat and the duration of its action are compared, it may be allowed to doubt that the germs in the vessels are all destroyed. It appears that micrococci and the so-called spores of *Bacillus subtilis* and *Bacillus anthracis* retain their vitality in considerably higher temperatures than do the fully developed Bacillus and shorter rod-forms.

It is true that the concentration of the fluid is changed if heated longer than to ebullition before sealing, and therefore one may entertain doubts of complete sterilisation so long as Dr. Bastian cannot either eliminate this possibility or disregard the change. One is strongly reminded of Needham's objections to Spallanzani's experiments in the last century, when Dr. Bastian says that if Pasteur's modification to continue the boiling were introduced, the urine might undergo a chemical change. But not alone is the uncertainty of a thorough sterilisation of great weight in the *à priori* estimation of the final success of such

experiments. The amount of oxygen remaining in the fluid is also considered by different authors a factor of greater or less moment with regard to the different forms of organisms or their earlier stages. The more recent investigations on this subject made known since the first publication of the present communication, as well as those of the best authorities previously, have shown that as the different forms retain their vitality with varied tenacity under the influence of higher and lower degrees of heat, so many are more or less independent of the amount of oxygen present. It may be freely admitted that the greater part of the gas is well driven out of the retorts, but some may still remain, and if so-called germs and lower forms are yet within the fluid or adherent to the inner surface of the retorts or to the exterior of the liquor potassæ tubes, they will find oxygen enough for their development. The unfavorable conditions into which such germs are brought by the treatment of the vessels and their contents, may well limit the degree of fertility ; on the other hand, as Dr. Bastian himself has found, the incubator temperature of 122° F. (40—45° C.) is a very favorable agent.

Notwithstanding the justice of these objections, which could be multiplied by an extended and a detailed reference to the Bacterium literature, what would appear as most remarkable in Dr. Bastian's numerous repetitions of the experiment, is the uniformity of the difference between the contents of the retorts with urine and unbroken tubes and of those with the mixed fluids produced by breaking the potash tubes within the retorts after closure and ebullition. The critics of Dr. Bastian have singularly enough not laid the proper stress upon this, the real point at issue, for even if all their objections were just, the fact of such a difference would be all the more interesting. In their trials of the method they have not scrupulously followed his directions, and he is well justified in refusing to accept their adverse statements for this if for no other reason.

Before entering upon my own experimentation I would with a word call attention to the apparent unchanged condition of the fluid in the one set of retorts. I have found, and the same observation has been made by others, that the degree of turbidity is not in direct proportion to the multiplication of Bacteria. Certain specimens of perfectly clear and apparently unchanged fluids may swarm with organisms, whilst in turbid urine with much sediment few if any organisms can with absolute certainty be discovered with the microscope. This, it is true, is not the rule, but as it may occur the fact of an apparently unchanged appearance alone is not a sufficient test. It appears that the formation of so-called zooglœa masses is in most cases the cause of turbidity.

The fresh filtered urine which I used never showed precipitation of phosphates or albumen on boiling, and in order to ascertain the degree of aoidity a previously boiled specimen of it was tested. This latter precaution, and the addition of the two-thirds proportion of liq. pot., was only adopted, however, after the result was found to be other than that recorded by Mr. Bastian, with urine whose degree of acidity had been determined with portions previously unboiled and mixed in the prepared retorts with liquor potassæ of the proportion of three-fourths of its acidity. To facilitate the determination of the degree of acidity a burette was constructed, with a capillary termination by means of which drops of known weight (0·0075 grms.) of the liquor potassæ could be mixed gradually with 30 c. c. of urine. The equality of the drops was insured by suitable precautions, the liq. pot. changed and the instrument well cleaned from time to time. In the first series of experiments litmus paper was used, but ever after rosolic acid, a very delicate reagent. In determining whether or not the contents of the retorts contained organisms, three to five specimens from every vessel were examined, and only then was their presence noted when they could either be distinguished by their characteristic movements or other signs of life, such as, for instance, an increase in the size of the zooglœa masses, and when caustic, liq. potassæ, and strong acetic acid failed to dissolve the suspected bodies. In all more than fifty retorts were charged and examined.

The fluids, which were mixed with the proportionate amount of liq. pot., and which were examined at intervals between twenty-four to seventy-two hours and fourteen days, were all found turbid and showed sedimentation, but *not one of them contained organisms;* the sediment was, moreover, found to consist of phosphates, crystalline, and amorphous masses on being examined by means of micro-chemical tests. Their odour was not appreciably changed, not even in those specimens which contained triple phosphate crystals.

This result remained the same after using urine from other individuals, and after adding peptone to some specimens. In order to make quite sure that organisms were not contained in the fluid, at least within the limits of all means of examination, portions of the same were allowed to form sediment in narrow glass tubes with a capillary termination, and the first drops carefully searched through under the microscope. In all these trials the result remained the same.

After remaining in the incubator during one month the specimens containing unbroken liquor potassæ tubes were found clear with very few exceptions, and, when examined, contained

no Bacteria, the sediment dissolving after addition of acetic acid; the degree of acidity had been in all such cases remarkably low.

I could not so far corroborate the statements of Mr. Bastian, and was unable to explain this remarkable divergence from the expected result. The disappointment was the more disheartening as the repetition of his experiments were made from the beginning more with a view to an explanation than as test experiments designed to determine the truth of his statement of fact.

There still remained seventeen retorts in the incubator, of which some contained mixed and others unmixed fluids. They were opened and their contents examined after they had been exposed to a temperature of 120° F. (40°—45° C.) between 65 to 126 days. They all contained more or less flocculent suspensions and a whitish-yellow sediment, which adhered to the walls of the retort; their reaction with rosolic acid proved to be alkaline and several smelled of ammonia. One only had a putrid odour, and was registered as not entirely without blemish. On addition of acetic acid the greater part of the sediment, which proved to consist chiefly of amorphous masses and of few crystals, was dissolved; here and there were found peculiar zoogloea-like bodies of yellowish tinge and a remarkable power of resistance to reagents, the exact nature of which I was unable to determine, and which frequently appeared in a later series of experiments; and lastly, *every specimen of urine, with and without admixture of liq. pot., and such also with perfectly clear contents, together with one whose fluid showed acid reaction, contained innumerable organisms of various forms.*

Micrococci and small spheroid bodies were most numerous, and appeared either as individuals or in torula- and zoogloea-form; together with these, and next in number, smaller rods (vibrio), single and in biscuit form, and lastly, variable quantity of bacilli of all lengths, free or articulated. These organisms showed the characteristic movements, or were grouped together in more or less dense masses. In no case was the degree of turbidity in direct proportion to the number of organisms, but it seemed to stand in relation to the zoogloea after the action of acetic acid had dissolved the other suspensions. It follows from this that the long-continued action of the incubator temperature is favorable to the multiplication of Bacteria, even when the remaining conditions introduced tend to retard their development. In what manner, if in any, the presence of the alkali acts as a factor was not thus far apparent, especially as organisms appeared where it was not allowed to mix with the pabulum just as freely as where it was added.

The next step was, therefore, to ascertain if there existed a causal connection or simply a concomitance of the phenomena. For this object the following elementary experiments were made. Fresh-filtered urine was put into test tubes, their opening closed with cotton and dipped into the water bath of 122° F. (40—45° C.). After thirty hours the reaction was alkaline, ammonia was generated, and countless Bacteria found in it. After some time the Bacteria sank to the bottom, and the urine was again clear and limpid. In spite of the lifeless condition of the organisms ammonia was constantly generated, and the degree of alkalescence enhanced. In order to determine the latter I made use of a burette, constructed as that for liquor potassæ, and added sulphuric-acid solution (5 per cent.), drop by drop, to a certain quantity of the urine, to which a small quantity of the above-mentioned alcoholic solution of rosolic acid was added. The change from a beautiful rose-red tint to a bright straw-like yellow, indicated that the first drop of acid in excess had been added. Perhaps this simple experiment goes far to show that the decomposition of urea is not so directly dependent upon living organisms as the vitalistic theory implies; but, on the contrary, that the continued action of the temperature of 122° F. (40—45° C.) is a favorable circumstance, if not an etiological factor of the same. Nor can the urea ferment of Musculus be its cause, for its action is said by the discoverer to be neutralised in 80° C. and by alkalies, both of which are made to act upon the contents of the retorts in Dr. Bastian's experiment. It is well known that urea is decomposed when its aqueous solution is heated in sealed glass tubes, or when in the dry state it is melted with an alkali hydrate. Mindful of the latter fact, I charged four test tubes with an aqueous solution of urea, and added to two of these a small quantity of dilute liquor potassæ. Two tubes, one with urea and the other with urea and the alkali, were sealed in the flame and placed in the incubator at 122° F. (40—45° C.). The remaining pair was dipped into the water bath of the same temperature, and closed with a cotton plug. The result was :

I. Test tube with urea and liquor potassæ, cotton plug. Generation of ammonia after seventeen hours in water bath.

II. As I, but without alkali. Feeble generation of ammonia, alkaline reaction after 139 hours in water bath.

III. Test tube with urea and liq. pot., sealed. Feeble generation of ammonia after twelve days in incubator.

IV. As III, but without alkali, was opened after fifty-seven days. Generation of ammonia not distinctly determinable, but

alkaline reaction was shewn with rosolic acid, whereas litmus paper was not changed.

It is clear, therefore, that at the temperature of the incubator and of the water bath urea is decomposed in a comparatively short space of time, and that the presence of the alkali serves to facilitate it. The slowness of the generation of ammonia in tube IV is of especial interest when brought in relation with the contents of the retorts of the first series. Here, also, when other tests for free ammonia are not available, a little of the solution of rosolic acid on a cotton plug, held over the mouth of the vessel, is a delicate reagent; care must naturally be taken that the plug does not touch the tube, nor that it be exposed to the air too long, for the smallest traces of atmospheric ammonia will show the colour reaction.

With reference to the change of the reaction, attention ought to be called to one retort, mentioned above, which contained organisms, and, notwithstanding, showed acid reaction. This retort was charged, July 11th, with urine of the acidity of eighty drops, together with a liq. pot. tube, which remained intact. On October 17th it was opened and examined, when the acidity was found to be reduced to thirty-five drops of 5·85 per cent. liq. pot. Dr. Bastian has already called attention to the reduction of the degree of acidity under similar conditions.

Notwithstanding the minute description which Dr. Bastian has given of his method it is, as in all cases of experimentation with fluids of unknown or inconstant composition, never possible to repeat the tests and draw the same conclusions. The simpler the pabulum, and the more we have it in our power to modify its composition, the more uniform will be the results of cultivations of organisms. I have, therefore, sought to study the question, making use of Mayer's solution,[1] modifying it in such a manner that, instead of adding tartrate of ammonia, I introduced urea (1·06 grm. to 30 c. c.), after the solution of the other salts had been boiled and filtered. After dissolving the urea the solution was again filtered. Although Cohn has found that this is not a very good pabulum, I would, in reference to my results, call attention to the fact that he himself has seen Bacteria develop and multiply in a simple solution of the milk sugar of commerce, and that he believes that the nitrogen was absorbed from the surrounding atmosphere. With this solution a number of test tubes were charged, sealed, and placed in the incubator 122° F. (40—45° C.). After sixty-five days all specimens were turbid and swarmed with organisms; the degree of acidity of 30 c. c. was reduced from 110 drops to 66 drops. It seemed

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[1] ·1 grm. of potassium phosphate, ·1 grm. crystallised magnesium sulphate, ·01 grm. tribasic calcium phosphate to 100 c. c. distilled water.

that the solution was too strongly acid, and the generation of ammonia too slow in order to change the reaction. In the next series a much weaker solution was employed. Retorts charged with the same, and prepared in the manner of Mr. Bastian's urine retort, *i. e.* with and without liq. pot., in proportion to the degree of acidity. Here, also, the result was similar to that in the case of the urine experiment. After ten to fourteen days the solutions mixed with liq. pot., and after fifty-seven days those with unbroken tubes all contained innumerable organisms.

As regards the forms of Bacteria which I found in all the above-described series of experiments, the greatest number were micrococci (Billroth), both in their characteristic movements as separate individuals, or linked in varying numbers and at rest, singly or in zooglœa colonies. The rods and bacilli were not so numerous; their contents appeared to be granular. In several specimens I also found forms much resembling ascococcus (Billroth). Here also I met with the zooglœa-like masses already mentioned, but am unable to determine whether or not they were organisms, as they showed no kind of movement, nor could I observe any multiplication of their elements.

It still remains to be proven that the multiplication of organisms in these fluids, treated by high temperature, and in a minimum of free and absorbed oxygen, is causally connected with the decomposition of urea and the generation of ammonia, which here seems to be the source for nitrogen.

The change in the following series of retorts, kept in the incubator during forty-one days, will conclusively demonstrate that ammonia takes such a rôle :

I. In dilute Mayer's solution and urea ($3 \cdot 0$ grms., 200 c. c.). The fluid remained clear, reaction alkaline, contained micrococci, spheroids, rods, and bacilli.

II. In dilute Mayer's solution and urea, with forty drops liq. pot. ($5 \cdot 85$ per cent.); little sedimentation, fluid clear, reaction alkaline, few moving micrococci and spheroids, together with crystals and amorphous, granular masses. It appears here as if the process of multiplication had already passed over.

III. In dilute Mayer's solution with dilute solution of caustic ammonia, which was introduced in the manner as the liq. pot. tubes in the first series, and then broken. Strong and diffuse turbidity, reaction alkaline, immovable organisms (micrococci, spheroidal forms, rods, and bacilli).

IV. In dilute Mayer's solution and urea, and a tube containing fifty drops of sulphuric acid (5 per cent.), which was introduced and broken as above. Fluid clear, low degree of acidity, here and there a micrococcus.

Finally, a portion of the contents of retort IV was divided and

put into two test tubes, into one of which a stream of air charged with ammonia was led, until the alkaline litmus reaction was obtained, the other remaining intact. Both were then closed by means of a cotton plug and examined after forty days. The contents of the second tube were in appearance and reaction unchanged, whereas the first was turbid and contained much zoogloea, and literally swarmed with different forms of organisms. It is worth noting that litmus paper indicated neutral reaction, but rosolic acid solution gave a very strong alkaline reaction.

A review of the above-described experiments shows, as regards Dr. Bastian's work, that in urine and the modified Mayer's solution containing urea, micro-organisms are developed or multiply after exposure to 212° F. (100° C.) for a certain length of time, after evacuating the greater part of oxygen and placing the retorts in an incubating temperature 122° F. (40—45° C.). But in the series here recorded this effect was produced only after a comparatively long time. It has also been found that it is not safe to draw conclusions regarding the presence of organisms in fluids from their outward appearance, i. e. the clearness or turbidty of the same, as the suspensions and sediments are frequently composed of other bodies, and as perfectly clear fluids often contain numberless organisms. Dr. Bastian found that micro-organisms appeared alone in such of the retorts as held urine mixed with liquor potassæ, and that after eighteen to thirty-six hours, whilst the urine unmixed invariably remained apparently unaltered. My repetitions of his experiments have led to a different result; in all specimens, irrespective of the admixture of liq. pot., Bacteria were developed, but only after a much longer sojourn in the incubator. The fact that they appeared sooner where the alkali had been added is explained without difficulty by the accelerated decomposition of urea and generation of ammonia, which provided the necessary nitrogen; in so far, it is true, liquor potassæ acts as a favorable factor.

As long as a doubt can be entertained regarding a perfect sterilisation, so long is it inadmissible to make use of these results as arguments in favour of "abiogenesis." I am inclined to consider these researches of some interest, inasmuch as the retardation of a development of lower forms or so-called germs which may have remained in the vessels can be naturally explained by the high temperatures and exclusion of free oxygen, whilst the multiplication of organisms can be understood when the incubator temperature and the generation of a simple nitrogenous body are considered as favorable factors. Urea may not, according to Cohn, be such a body, but when ammonia is generated in presence of the necessary inorganic substances Bacteria can multiply even without an unlimited supply of free oxygen.

II.

With a view of arriving at a more definite knowledge of the effects of an addition of ammonia to a solution of salts free from nitrogen, I proposed next to study its influence under the microscope, introducing this method by the following more elementary series of experiments.

A number of retorts were prepared like those of Dr. Bastian, being charged with Mayer's salts solution, minus the vehicle for nitrogen, which latter was represented by twenty drops in each of a dilute solution of ammonia (ten drops caustic ammonia, 100 c. c. distilled water) in sealed tubes, which were afterwards broken and their contents mixed with those of the sealed retorts. The result proved to be, in numerous repetitions of the same procedure, that after remaining about seven days in the temperature of the laboratory the fluid contained numberless organisms of various forms.

Continued observation under the microscope was facilitated by a simple chamber consisting of a glass slide, upon which was fastened by means of Canada balsam a square of hard rubber with a central boring measuring 4 mm. in thickness.   On each of two opposite sides a fine metal tube was let into the square in such a manner that a stream of air could pass through the chamber.   A drop of the salts solution, minus nitrogen, having been brought on a thin cover glass, which was held down by a thin film of pure olive oil on the upper surface of the rubber square, the field of observation was now in position, the drop hanging in the chamber, on the bottom of which a drop of distilled water presented evaporations.   A rubber tube of small calibre is drawn over one of the metal tubes and connected with a Woulff's flask in such a manner that a stream of ammoniated atmospheric air can be gently forced into the chamber.   In order to prevent aspiration when the afferent rubber tube is closed, a short piece of rubber tubing is attached to the opposite metal tube, whose distal end, provided with a glass mouth piece, is kept plunged under water.

With this simple apparatus, which can easily be kept scrupulously clean and can be taken apart when necessary, the development of micro-organisms and its modification when ammonia is provided or withheld can be observed for many days in succession.   Here, also, a small cotton plug soaked with a little rosolic acid and applied where the current leaves the chamber is of value in determining whether or not ammonia passes through the chamber.

The results of my observations have led me to believe that the first stage in the development of micrococci, rods, and bacilli, as

well as of glœo-coccus or glœo-bacteria (Billroth) is represented either by what could only be distinguished as an extremely small speck in the field or of spheroidal bodies much resembling small fat-globules in appearance, but not in their chemical nature, so far as could be determined by means of chemical reagents. In one case the smallest speck assumes lively motion, in another it remains where first seen, and little by little a conglomeration is formed of many like bodies ; I was unable to determine whether this was a process of division. In another part of the field a spheroidal body is gradually elongated, and finally a bacillus is found in its stead, and presently a thin septum is found to part it into two individuals, and so on until the chain of bacilli extends in some cases from one end of the field to the other. When the zooglœa colonies have reached a certain size they sink to the bottom of the drop. I have found that in those parts nearest the under surface of the cover glass, micrococci and zooglœa generally develop most abundantly, whereas bacilli are most numerous in those parts of the drop nearer to the atmosphere of the chamber. Other globular bodies send out processes of comparatively considerable thickness, which are oftentimes drawn in repeatedly or after assuming a knob-like shape, at their extremity are gradually separated from the mother cell ; this mode of prolification closely resembles that of yeast cells, but careful measurements have shown that the organisms in question are smaller than Torulæ.

When the rubber tube was compressed during seventeen hours, and in this manner ammonia and a fresh supply of atmospheric air was withheld, no bacilli were to be seen in the fluid, only micrococci globules and short rods ; but after allowing the ammoniated air free access, and gently forcing it through the chamber, bacilli appeared after twenty-four hours ; the movements of the micrococci and of the rods wholly ceased, however, when the metallic clamp was allowed to compress the afferent hose for a longer period of time. The movements of the bacilli and of the rods were such that the body proper remains unbent ; it is, therefore, very natural to suppose that they are supplied with a ciliary appendix ; I was, however, less fortunate than several other investigators, and could not discover them. When a second Woulff's flask, containing distilled water with a small known quantity of carbolic, acetic, hydrochloric acid or camphor was connected with the apparatus, so that the current of air passed through these liquids before reaching the chamber, the development of Bacteria was seen to begin only after at least five days, and only a few bacilli appeared within a week. During this time the current was made to pass gently through the chamber every two hours in the daytime, just as in the

operation with ammoniated air above mentioned. Here the rosolic-acid cotton plug was not reddened, no free ammonia reaching the chambers. As a control experiment, the same fluids were added to solutions contained in test tubes, and which swarmed with organisms. No diminution in the violent movements of the Bacteria ensued, nor was their number apparently diminished. It is clear, therefore, that the vapours of these so-called antiseptic agents were not the cause of the retardation in the development or multiplication of organisms. That the organisms appear at all, but only comparatively late, is easily understood when it is borne in mind that Cohn observed in a simple solution of milk sugar a multiplication of Bacteria which derived their nitrogen, according to his hypothesis, from the atmosphere, to which fact I have already called attention, whilst Schönbein found that when water evaporates in atmospheric air nitrogen is made free.

The success in practice of the treatment of wounds according to Mr. Lister's method is now very generally admitted, but the hypothesis on which this method is based is not so universally accepted. My experiments are, in this respect, of some interest, but I am not willing to claim for their results any very general application. Observers of high rank still assert, after careful researches, the "ubiquity" of organic germs, and it is clear that such a view cannot be logically made to harmonise with the views held by Mr. Lister and his more ardent disciples.

The antiseptic properties of the substances which I have used being conceded, I hold that they are harmful to the development and multiplication of organisms from the readiness with which they unite with substances indispensable to the existence of the organisms rather than on account of their direct antizymotic properties. I freely grant that these researches do not conclusively prove this supposition, but they seem to indicate such a relation.

Some TEACHINGS *of* DEVELOPMENT.   By E. A. SCHÄFER, F.R.S.,
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I.

DEVELOPMENT is the term applied to the process of evolution of
an organism from its simplest to its most complex phase of
existence.

In all but the very lowest animals the development of the
individual begins with the ovum or egg, this being the elemental
portion or cell of the parent animal which is set aside for the
special purpose of reproduction.   In most cases the separated
cell is incapable by itself of reproducing the parent ; it must first
blend with a separated part of another parent.   This blending
with the ovum (female reproductive element) of the anthero-
zoid (male reproductive element) constitutes the process of
fertilisation.

The commencement of development follows immediately upon
the completion of fertilisation.   Once commenced the pr cess is
usually continuous, and being accompanied by a general increase
in size, the result is the perfect or adult individual.   Although
these two processes (development, *i. e.* increase of structural and
functional complexity, and growth, *i. e.* increase of size) generally
go hand in hand, we must be careful to distinguish between
them.   For mere increase of size may often give the appearance
of greater complexity, when in reality the organism has not
advanced either in function or in intimate structure.   This is
the case sometimes when the increase of size takes the form of
budded outgrowths which are merely repetitions of the structure
of the parent organism.   The common freshwater polyp con-
stantly presents us with an illustration of this, for its numerous
buds produce an appearance of complexity which disappears
when the buds detach themselves as independent organisms.   In
the compound hydroid polyps, in which the buds remain attached,
the appearance of complexity remains throughout the life of the
organism, and is accompanied in some by an actual progress in
functional and structural complexity.

In the case just alluded to the increase of size accompanying
development takes the form of an arborescent repetition of all the
chief parts of which the developing individual is composed.   But
it may instead take the form of a serial repetition such as we see

[1] This article contains the substance of the last two of a series of
twelve lectures on Animal Development, delivered at the Royal Insti-
tution in Jan., Feb., March, 1879 ; and has not been modified since that
date.

in the Strobila stage of development of the larger Medusæ, and which presents itself in striking contrast to the arborescent repetition of the corresponding stage of development of the Craspedote Medusæ. In the so-called Strobila the developing individual has become partially separated by transverse constrictions into a series of perfectly similar, saucer-shaped segments, each of which comprises within itself in a rudimentary form, a representative of all the parts of the originally simple parent organism. So long as these remain united the Strobila is a compound individual constituted by a serial repetition of similar parts ; but they soon prove their individuality by breaking away from the compound body and from one another, and maintaining existence as independent organisms.

This serial repetition is seen in still more characteristic form amongst the Annelids, where almost every one of the many segments which compose the body is complete within itself, and any one might be taken as a representation of the primarily simple embryo which produced them all. And viewing them by the light of the illustrations furnished us by the Cœlenterata, it seems not unreasonable to regard all animals in which such serial repetition is found as compound rather than as simple individuals—to a certain extent comparable to the condition of the united polyps just referred to, but far more closely joined together and interdependent ; rendered thereby incapable of maintaining themselves as separate organisms, but, on the other hand, able the better, by their united powers, to carry on the struggle for existence. And the importance of this serial repetition is indicated by the fact that it is found as the main characteristic of the two most highly organised branches of the animal tree—the Arthropoda and the Vertebrata, masked though it often is in them by that tendency to recoalescence which at first sight seems a recurrence to more simple types of existence, but which, when attentively considered, proves to be merely the expression of the closer union and combination of the separate segments, the better to work together for special objects or for their mutual benefit.

I have stated that, once commenced, the process of development of an animal is a continuous one. Nevertheless, it is customary to regard it as advancing by separate steps or stages —to look upon it as a staircase or ladder, the stairs or rungs of which are successively attained, and the number of which depends upon the complexity of organisation of the adult individual. The familiar phrases " scale of development," " scale of organisation" (Latin, *scalæ*, a staircase), are a sufficient indication of the mode in which the subject is generally viewed. And although I think that the continuous nature of the process

should on no account be lost sight of, yet it is so manifestly convenient for purposes of description and of comparison to maintain the idea of grades, that I shall here adopt a like simile, choosing a succession of telescopic tubes, instead of a staircase, to represent the succession of developmental phases, because those can be imagined to be more or less drawn out, and shifted upon one another, so as to illustrate the varying relations which the typical successive phases bear to one another in the development of different animals. It will further be necessary to imagine the tubes constituted of some very elastic material, rendering it possible for them to be extended to two, three, or many times their proper length, or, on the other hand, to become shortened so much as to be scarcely perceptible. Each tube of which the telescope is composed may be taken to represent a particular stage of development, and the greater the complexity of the organism the more tubes must be added to the series, and the greater the length of the developmental telescope.

If we now proceed to compare the development of various animals with one another, we are struck with the resemblance—with the identity I might rather say—of the first stages in all. To begin with all take origin in an ovum; and although at the first glance this appears so different in many (compare the ovum of a mammal with the egg-yolk or ovum of the bird), examination shows it to be in all cases similar in essential composition. For the difference depends solely upon the amount of nutritive material which has been stored up within and in connection with the reproductive protoplasm, and has relation to the greater or less facility with which a supply of food can be obtained from without during the active developmental process. To use a familiar comparison, the difference is precisely the same as that between an army operating in a fertile country where supplies are readily obtainable, and a force which is to operate in a region barren or devastated, and far removed from the base of supplies. In the one case pabulum sufficient merely for immediate use is required, in the other a great store of nutriment must be accumulated and must accompany and proportionately hamper every movement of the army. Moreover, since to a part of the force must be entrusted the duty of guarding the store and distributing it as required, a greater number of men is required to bring the army up to the same proportionate strength as that of the unencumbered force. So in those ova which are provided with a large store of nutritive material, do we find that when the active movements which characterse development commence, those movements are hampered by the presence of the inert food-material, and it is precisely as the store becomes lessened that the apparent difference between such ova as these, and those others

which from the first have scarcely any store of nutriment, disappears. The difference is one of degree only and not a difference in the essential nature of the ova; this view is confirmed by the existence of every shade of transition between them, as well as by observing that the difference obtains in the ova of animals which are otherwise closely allied.

Further, in the ovum of all animals there follows immediately on fertilisation the successive division or cleavage of the single cell into first two, then four, and so on until an indefinite number of cells is formed which arrange themselves around a central space —the cleavage-cavity. This, in the majority of cases, is distinct enough, being occupied merely by clear fluid and enclosed by a single layer of cells; but in some instances it is partially or wholly filled up by the excess of food-material and its immediately attendant protoplasm, which may or may not, according to its relative bulk, have participated in the active changes which have been occurring in the rest of the protoplasm, and of which the cleavage is the result. To revert to our military comparison it is as if the two armies had broken up into detachments and arranged themselves so as to enclose circular areas of ground; in the one case the detachments unencumbered and active on all sides, with the enclosed area left clear; in the other case the *matériel*, with the camp followers, packed away in the centre and partly occupying one side of the space, whilst the least encumbered detachments are chiefly accumulated on the side where most activity is required.

Bearing in mind this difference, and regarding it as accidental, we may take the completion of the cleavage process, as far as the arrangement of the resulting cells around a central area in the manner described, as the first stage in the development of all animals. It may be termed the stage of the *unilaminar blastoderm*, or the *blastosphere*. For the name blastoderm has long been applied to the cellular membranes of which the developing ovum is formed at early periods; and the term blastosphere may be used, because at this stage a typical ovum, *i.e.* one with a minimal amount of nutritive material, consists as we have seen of but a single uniform layer of cells forming the wall of a hollow sphere.

We meet with this typical condition of the first stage in Gastrophysema (according to Haeckel), in Phallusia, and in Amphioxus and other animals. But even in those ova which contain only a slight excess of pabulum we find that there are differences observable in the cells which form the wall of the blastosphere, for the food-material becomes chiefly collected at one part of the layer, rendering the cells at this part more granular and larger than the others. This difference may be so

slight as to be almost imperceptible, as in the Holothurian blastosphere, or it may be more obvious as in that of the Sponge and Paludina. And every transition is found between these cases and those in which the amount of pabulum is so great in proportion that it can no longer be contained in the typical single layer of cells, but becomes accumulated in the cavity of the sphere as well, either still enclosed in definite cells as in the Amphibian, or for the most part unenclosed in cells as in the Fish, the Bird, and the Crustacean.

The next most important change that occurs in the typical unencumbered ovum is the invagination of one part of the simple wall of the blastosphere, in such a manner as to convert the hollow single-layered vesicle into a cup with double walls. The cleavage-cavity or cavity of the blastosphere is concomitantly reduced in size or even obliterated as it is encroached upon by the invaginated part of the wall, and its place is taken by the cavity of the cup. The orifice of this, at first widely open, becomes gradually closed up by the continued growth of the two layers at the mouth of the cup, until but a narrow aperture remains, which persists for a time, but at length in many cases becomes closed. This aperture is termed by Haeckel the primitive mouth or *protostoma*, by Lankester the *blastopore*.

The result of the completion of this process of invagination is again a hollow vesicle, but its walls are now composed of two layers of cells instead of one. The inner or invaginated layer is termed "entoderm" and the outer or enclosing layer "ectoderm." The cavity which they enclose always becomes the future alimentary cavity, and hence the name "gastrula" has been applied by Haeckel to this stage of development. We may also speak of it as the *cup-stage* or as the stage of the *bilaminar blastoderm.*

In typical ova the original blastosphere is uniform throughout, and it would be impossible to point out the part of the wall which is to be invaginated. But in those ova which have even a small excess of nutritive material and in which therefore, as already mentioned, some of the cells of the blastosphere are characterised by containing this excess, we always find that it is this particular part of the wall of the vesicle which is involuted to form the inner layer of the cup. So that even in the stage of the blastosphere we can predict which cells are to become entoderm and which ectoderm.

Finally, in those ova in which the nutritive material largely preponderates we find that even when the cavity of the blastosphere is not entirely filled up by that material (and of course also when it is so filled up), the mechanical hindrance to any invagination is so great that the process cannot be effected.

Still, even in such cases as these the cup-stage is not absent.
The same result is obtained, but in a different way.   For the
enclosure of the pabulum-containing entoderm by the ectoderm
is effected solely by the growth of the latter, without any accom-
panying invagination of entoderm.   In these cases the cavity of
the gastrula also is necessarily occupied by the pabulum, and
this if greatly in excess may even project beyond the cup-orifice.

We may then take the gastrula or cup-phase as a second stage
and note that, like the first, it is met with (modified only by the
accidental presence of food material) in all animals above the
Protozoa.

There are two apparent exceptions to the general rule of for-
mation of the gastrula stage by invagination.   These are met
with in the development of the freshwater polyp (Hydra) as
described by Kleinenberg, and that of one of the Medusæ (Ger-
yonia) as described by Metschnikoff and by Fol.   In these
animals the bilaminar blastoderm is said to be formed by the
splitting into two portions, inner and outer, of some or all of the
cells of the single layer which forms the wall of the blastosphere,
the inner parts becoming collectively the entoderm, the outer
remaining as ectoderm.   Regarding these exceptions the ques-
tion naturally suggests itself, Are they due to defects of obser-
vation ? and further, supposing that there is no flaw in the facts,
are they nevertheless explicable as modifications only in the
ordinary mode of formation of the gastrula ?   Certainly as far as
concerns Hydra, the recorded observations of Kleinenberg are
too incomplete on this part of the development for his conclu-
sions to be accepted without demur.   And it is very generally
admitted that the development of Geryonia needs careful rein-
vestigation with the aid of microscopic sections.   It is not
improbable that the result of such reinvestigation might prove
the existence of some sort of invagination at a much earlier
period than that at which the so-called delamination or splitting
occurs.

The cavity of the cup at first communicates, as we have seen,
with the exterior, by means of the aperture of invagination or
protostome, and when this becomes closed, or even before its
closure, another aperture appears, and in most cases yet another.
These secondary openings into the cavity of the gastrula become
respectively the anterior and posterior orifices of the alimentary
canal, and their formation is always accompanied by an ingrowth
of ectoderm towards the endoderm.   One or other of these aper-
tures may be formed in the place which was before occupied by
the now obliterated protostome, or in some cases the latter may
itself remain persistent as one of the secondary orifices.

We may now pass to the consideration of the third essential

forward step in development. This is the separation or segre-
gation of some cells from either one or both primary layers to form
an intermediate set, which acquires greater proportional import-
ance the further upwards we trace it in the scale of organisation.
Since the cells of this intermediate set in most cases, and certainly
in typical ova, are not of independent origin, but are derived in-
directly from one or other of the two primary layers, it is clearly
a mistake to regard the layer which they form as of equal im-
portance with the other two—an idea which is distinctly implied
by the name " mesoderm" usually given to it. The two primary
layers—the "foundation membranes" of Huxley ; "ectoderm
and endoderm" of Allmann—are distinct and well defined. The
so-called mesoderm is often very different in these respects, so
much so that in some animals parts which are looked upon as
mesodermic by some morphologists are regarded as still belonging
to the two primary layers by others.

This mistake of regarding the mesoderm as of equal morpho-
logical importance with ectoderm and entoderm has originated
in the study of the development of the higher animals, in which,
in many cases, the relative time of its origin has become shifted,
so that it may begin to appear almost simultaneously with the
primary layers. And even in some animals, not very high in the
scale, the same shifting may be found. Thus, for example, in
the Holothurian, the separation of some of the ectoderm cells to
form part of the intermediate or mesodermic set has already
begun, even before the commencement of invagination, and, there-
fore, whilst the general development has not advanced beyond
the blastophere stage. And the same shifting is carried to a
still greater extent in Clepsine, where we find that even when the
cleavage of the ovum has advanced but a step or two, certain
portions are separated to produce, as the cleavage process con-
tinues in them, the whole of the mesoderm. In this case,
then, the third stage has begun almost as soon as the first stage
itself.

This premature separation or precocious segregation (Lan-
kester) of parts which analogy would lead us to expect later is
a very common feature in animal development. It is illustrated
in Clepsine in a still more remarkable way, in the premature
separation of the portions of the dividing ovum from which the
cells of the nervous system are derived. And it is on account of
this tendency which the phases of development exhibit to become
shifted to periods earlier than usual for their appearance that I
have compared them to the tubes of a telescope, for we can so
much the better illustrate them in their relations to one another.
Thus, in the case of the Holothurian, we should shift the third
tube representing the formation of the mesoderm downwards, so

as to overlap the second one representing the gastrula, and in the case of Clepsine so as almost to reach the base upon which the telescope rests.

II.

At the end of the previous lecture we had arrived at the consideration of what we, for convenience, regarded as the third stage in the developmental staircase, the third tube in the developmental telescope—the formation, namely, of the mesoderm ; and I insisted that the term is a misnomer, because it obviously places itself side by side in the mind with the names which have long been applied to the two primary layers, "ectoderm" and "entoderm," whereas it is secondary to these, being derived from them. The mesoderm consists, in fact, of cells which have been separated or segregated from amongst the cells of the primary layers for the performance of special functions. In the lowest animals in which such a separation occurs it may take place at any part of the primary layers, or even over the whole of their extent. We see this in the sponges and jelly-fish. In these, when intermediate cells are separated, they lie in a jelly-like substance, and the main purpose which the segregation subserves is that of support and connection. Now, it is obvious that this sustentacular function might be almost as well performed by the inert jelly alone—in fact, the soft protoplasm of the cells can be of but little assistance, so that we should naturally look upon the cells in this primitive mesoderm rather as ministering to the nutrition of the jelly than as agents in the performance of its function. This view is strengthened by observing that it is the intermediate jelly-like matrix of this primitive connective tissue which is the first to appear, the cells (when they do occur, for in many cases they are absent throughout life) wandering into it subsequently. In the larger Medusæ (Aurelia) they come, according to the testimony of most observers, from the entoderm ; in the Sponge they are derived from the outer of the two layers which are found in the Olynthus stage—the one which is generally regarded as ectoderm.[1]

[1] There seems to be some uncertainty about the interpretation of the two layers of cells which are found in the Olynthus stage of development of the Sponge. According to the descriptions of various observers, and especially that given by F. E. Schulze with regard to *Sycandra raphanus*, the segmentation of the ovum takes place, and a blastosphere becomes formed much in the usual manner. Of the cells of this blastosphere those of one hemisphere are smaller clearer, and provided with long cilia, those of the other and lesser hemisphere being larger, more granular, and without cilia. Presently the latter become invaginated, but the cupping thus produced proves a temporary condition merely ; fluid again accumulates in the cavity of the blastosphere and obliterates the cup-

Another proof, if one were needed, that the jelly is the primary and the cells which wander into it the secondary part of the purely sustentacular mesoderm of these lowly organised animals is to be met with in the fact that in some—*e. g.* Hydra and the smaller Jelly-fish—the jelly is the only part of the layer present. The nutrition of the jelly is administered directly by the entoderm cells.

We see then that in the lower forms the only function which is delegated to the intermediate layer is the mechanical one of support. But in all the higher animals the segregated mesoderm cells are deputed to perform other and more important functions as well, for, besides the connective and supporting structures of the body, the actively contractile tissues which are concerned in effecting the movements of the body are derived from them. In the Medusæ or jelly-fishes this function is still performed by a tissue which is undoubtedly part of the ectoderm, although at first sight it seems to constitute a distinct layer of cross-striped fibres beneath the ectodermal epithelium. For if these fibres are carefully investigated, it will be found that they are many of them placed in the interior of large cells which project to the

form. After a certain interval a more decided cupping again takes place, but this time it is the clear ciliated cells that are invaginated. This condition remains permanent. The cup-like sponge settles down and the orifice of the cup becomes closed; a jelly-like substance accumulates between the two layers of which the cup is formed, and cells pass into it from the outer layer; one or more canals become bored through the jelly so as to effect a communication between the cavity of the sponge and the external medium; and the sponge may be regarded as formed. Now it is generally assumed that the first cupping being temporary is a mere accident, and that the second represents the gastrula stage of other animals. If this is really the case there is of course no fault to find with the generally received interpretation which sets down the large cells of the ciliated chambers of a sponge as entoderm and all the rest of the sponge (viz. the flattened cells which cover the external surface and line the water-canals, and the general thickness of its jelly with its included branched cells), as ectoderm (and mesoderm). But it is difficult to avoid the thought that by ignoring the first cupping and setting it down as accidental or at least as unimportant on account of its transciency, an error may have been made and a divergent if not a retrograde process of development regarded as a normal and progressive one. It is admitted that the microscopic appearance of the ciliated hemisphere of the sponge-blastosphere is in favour of its being looked upon as ectoderm rather than entoderm, and conversely with regard to the non-ciliated hemisphere. The origin of the branched cells in the jelly from the non-ciliated cells is a fact pointing in the same direction. And although our knowledge of the physiology of the sponge is too imperfect to be of much service in deciding the question, nevertheless the vegetative mode of life which sponges exhibit would lead one to expect a proportionately increased entodermal and diminished ectodermal development.

If the conjecture thus sketched out is a sound one it follows that sponges, as compared with other animals, are turned inside out.

surface between the cells of the general layer of ectoderm, which covers the under surface of the umbrella, and they evidently belong to that layer.   In other words, the deeper parts of some of the ectoderm cells are modified to form the muscular structures.   In Hydra, according to the descriptions of Kleinenberg and Korotneff, this is still more clearly the case—in fact, almost all the large superficial cells of the ectoderm are thus modified in their deeper part, although the muscular development is less characteristic.   Now, supposing these muscular cells of the jelly-fish, in place of remaining amongst the rest of the ectodermal cells, to sink deeper into the surface of the jelly, or even to become imbedded in its substance, they would then appear altogether distinct ; and although originally derived from the ectoderm, would be reckoned as part of the mesoderm.   This is, indeed, the condition which is actually found in many other Cœlenterates.

We have seen that in the lower forms of the Metazoa the cells of the intermediate layer are derivable from almost any part of the primary layers.   But in all the higher forms the mesoderm is developed from one part only of those layers, and this part is very frequently close to the contracted orifice of the gastrula, at the place where ectoderm and entoderm pass round into one another.[1] And from this place of origin the mesodermic cells—consisting of those which are to minister to support, and those which are to minister to movement intermixed—spread out between the two primary layers.   We see a typical instance of this in Paludina and also in Unio.   Now, if we suppose that the segregation appeared first at one particular point at the margin of the protostome, and afterwards spread in all directions from that point, we can comprehend how bilateralism might have appeared as the result of the separation of mesoderm cells (accidentally ?) at one part only of the margin of the protostome. But whatever might have been the original conditions, the mesoderm as we now actually see its development, appears from the first in two halves, and bilateralism makes its appearance simultaneously with these.

Our third stage, then, such as we see it in all animals above the Cœlenterates,—the segregation, namely, of an intermediate

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[1] If we agree with Haeckel in surmising that in the earliest stage of animal evolution in which the cup-form or gastrula appeared, the orifice functioned as a mouth, we can readily understand how it is that a general mesodermic cell-segregation should have tended to localise itself in the neighbourhood of that aperture. For this would probably, earlier than other parts, become the seat of special active functions ; e.g. the opening and closing of the orifice or even the sucking in of aliment, such as we see occurring in the early embryo of the earth-worm.

set of cells from the entoderm and ectoderm, destined to sub-
serve the functions of support and motion, and termed " meso-
derm "—has in reality in all probability been produced by the
coincident occurrence of at least two distinct segregations. Per-
haps one of these, the muscular, is precocious, and has blended
with the other, the sustentacular, which should have preceded it.
If this is the case each of the two segregations should be re-
garded as constituting a distinct stage in development. But they
are so completely blended in their origin in the higher animals
that it is impossible to differentiate them.

There is no reason to suppose that the two layers into which
the mesoderm subsequently splits, in animals in which a body-
cavity, or cœlom becomes formed, have anything to do with this
supposed primary double segregation of the layer.   For the
segregated cells are entirely intermixed as the mesoderm spreads ;
although they may afterwards become again partially separated in
groups (for the constitution of muscles, cartilages, &c.), according
to the function for which they are destined.   The formation of
the cœlom is a distinct forward stage in development, and is the
first step in the direction of the formation of a circulatory
system.   The cells which bound both the cœlom and its offsets
are in most animals segregated from the general mesoderm, and
belong to the set of sustentacular cells, but in the Holothurian,
according to the description given by Selenka (and also in
Sagitta and Amphioxus as shown by Kowalewsky), they are
derived directly from the entoderm of the alimentary cavity, and
already before their severance from this, enclose a commencing
cœlom. There is not sufficient ground for regarding these lining
cells of the cœlom and vascular system as constituting a special
segregation.   In vertebrates they are certainly of the same
nature as those of the sustentacular part of the mesoderm (the
connective tissue), and the same is probably the case in the other
animals where they are found.

Another well-defined forward stage in development is the
occurrence of a special segregation of ectodermic cells for the
performance of the nervous function.   These differ from those
which are destined for the muscular function in that they are
never blended in their origin with the mesoderm, and indeed do
not in any animal lose their primitive connection with the
ectoderm until development is comparatively advanced, if even
they do so then.

As in the case of the muscular segregation the first indication of
a separation of some of the ectodermic cells for the performance
of nervous and special sensory functions is met with in the
Cœlenterata.   This takes the form of a prolongation of the
attached ends of some of the ectoderm cells into branched

fibres, which interlace with those of neighbouring cells, and probably serve to convey any impressions received by the cells of which they form a part, to deeper lying muscular cells which have lost their place in the superficial ectodermal layer and their connection with the external medium.

The "nerve-epithelium cells" thus formed may themselves tend to sink below the general surface of ectoderm. If this happens they lose the characteristic shape of epithelium cells, and become rounded, with extensions in the direction of the nerve-prolongations. In fact, like the muscular tissue, they assume a minute structure which is similar to that of the nerve-cells and nerves of the higher animals. Nevertheless these nerve-cells in the jelly-fishes in no case separate themselves from the ectodermal layer. They lie, in fact, between it and the muscular layer in those parts in which the latter occurs.

And the sense-organs in this branch of the animal kingdom also show that their essential place of origin is from the ecto-derm. For visual organs first make their appearance as patches of ectoderm-cells filled with coloured pigment, and some of them with nerve-fibre processes connecting them with adjacent nerve-epithelium cells; auditory organs as ectoderm-cells, con-taining crystals in their interior, and similarly connected; and olfactory organs as little pits lined by ciliated ectoderm-cells, and connected likewise to a nerve-epithelium.[1]

Amongst other animals we observe that in Sagitta also the segregation of ectoderm to form a nervous layer is in the first instance general and not localised. Eventually this segregation becomes accumulated mainly in two situations to form the cephalic and abdominal ganglia. In Amphibia, too, the separation of ectodermic-cells to form a nervo-sensory layer is at first general. But in most animals, e. g. the Earthworm, Euaxes, Ascidia, Amphioxus, the segregation in question begins at one part only of the edge of the protostome, as is the case with the meso-dermic segregation—this situation having been possibly deter-mined by the high functional importance of this orifice (assum-ing as before with Haeckel that it originally served as a mouth). Extending from this situation, the nervous separation would no doubt be chiefly guided by the arrangement of the pre-formed muscular segregation, and would hence tend to assume a bilateral condition such as we see it to possess.

[1] It is the opinion of Professor Claus that the depression which is seated above the base of each lithocyst in Aurelia and allied forms of Medusæ represents an olfactory organ. And the comparative researches of Drs. O. and R. Hertwig have rendered it more than probable that the otoliths of the jelly fish even when they appear, as in Aurelia, to be connected with entoderm alone, are originally derived from the ecto-derm.

We also find that cells which are set aside for the sole per-
formance of special functions invariably become modified in the
arrangement of their living substance, and in some cases also
in its chemical nature. The structural changes which thus
accompany and indicate the assumption of any special function
by cells, constitutes the science of histology. It would be
carrying us too far from our immediate subject to enter into a
detailed account of the nature of these structural changes here,
so I will only point out two facts with regard to them. One is
that the special modifications of structure which the cells assume,
that have been set aside for the sole performance of special
functions, are similar throughout the animal kingdom in all
essential features. The second is that in the lowest types
where they occur the structural characters appear before the
segregation of the cells is complete, whilst on the other hand
in the development of an individual belonging to the higher
types such segregation may have been long effected before either
the functions or the accompanying structural changes in the cells
begin to be manifest; another instance of premature segregation.

We have thus far been speaking of the separation of special
sets of the cells of which a simple organism is composed for
the performance of special duties. But in the united or com-
pound organisms, before considered, it very frequently happens
that one or more of the units of which the compound body is
composed becomes altogether specially modified for the perform-
ance of one duty to the exclusion of others. Thus we see that
some of the individual buds, of which the compound organism
of a hydroid polyp consists are adapted for purposes of prehen-
sion and alimentation, whilst others are adapted solely for
reproduction. This localisation of function in the different buds
of a compound organism is carried to an extreme degree in the
Siphonophora—animals which on the whole resemble the hydroid
polyps, but instead of being fixed to a rock at the bottom of
the sea, float near the surface of the water, looking like strings
of beautifully iridescent hyaline beads. In them we find the
specialisation of individuals to have proceeded so far that whilst
one of the innumerable individuals which have been produced by
budding from the original single one, is set aside to perform the
purely mechanical purpose of suspending the united colony near
the surface of the sea, and is changed into a minute balloon
enclosing a bubble of air; others are deputed to propel the
organism through the water and become wholly transformed into
so many pulsating bells; others again have merely to receive
tactile impressions from objects in the external medium, and are
chiefly metamorphosed into long feelers; others are occupied
solely with the seizure and ingestion of victims for the food of

the whole compound organism, and mainly consist of a stomach with a long stinging trailer appended ; others have confided to them the reproduction of the organism ; whilst all the more delicate of the thus modified individuals can shrink for shelter under certain other of the members of the colony which have degenerated into firm protecting scales. And in that higher type of compound organism which is characterised by serial instead of budding repetition, whole individual segments are often as distinctly set apart for the performance of a special function, although from their close union one with another it is generally difficult to trace such complete specialisation.   The extensively found adaptation for prehensile and masticatory purposes of one or more of the anterior segments in so many of these organisms readily comes to mind as an illustration of this.

But let us revert to the consideration of the sets of cells which become segregated for special purposes from the primary blastodermic layers.   Once formed they are found to proceed through stages of their own, varying in number with the extent of development of the organs which they compose in different animals. In one group of animals it will often be found that one or more such segregations have progressed in their functional, and correspondingly in their structural development much further than others.   In illustration of this we may notice that in the Vertebrata it is the nervous segregation, in the Arthropoda the muscular segregation, which above all others has attained the greatest development.   What mammal, for instance, is capable of the tenth part of the activity in proportion to its size, which is evinced by the sustained flight of the dragon-fly or the prodigious leap of the flea !

As an illustration of the progression in development of a special segregation, it will for our present purpose be most instructive if we trace out the early stages of formation of the nervous system in one of the highest types, and if we compare those stages with what we find in animals lower in the scale.

In the Toad, which we may select as a typical Vertebrate, there is at first a uniform layer of cells which is separated at an early period from the ectoderm ; then a thickening of this layer occurs on either side of the axis of the embryo, extending forwards from the protostome, the two thickenings forming the boundaries of a groove which lies between them ; we next find the groove becoming roofed over by upward extensions of ectoderm, both from behind the protostome and on either side, and thus converted into a canal which is at first open in front and communicates behind through the protostome with the alimentary cavity ; next (or even before the roofing in is completed) we observe that the anterior part of the nerve-tube becomes en-

larged and subdivided to form the primary parts of the brain, and finally, the wall of the tube becomes differentiated to form nerve-cells and nerve-fibres.

If now we compare these stages of development of the nervous system of the Toad with temporary or permanent conditions of the same system in certain animals lower in the scale, we are struck at once by the fact that the various stages described are more or less represented by those conditions.

Without again referring in detail to the mode of formation of the nervous system as a general segregation from the ectoderm, which is met with in the Medusæ and in Sagitta—for it might perhaps be accounted too bold to attempt a comparison between these and the earliest stage, that of general segregation, in the Toad—we find that in the Earth-worm the nervous system commences exactly as in the Toad, in the form of two thickenings of the ectoderm which extend forwards from the cup-orifice and form the boundaries of a shallow groove. But the development does not proceed further in the same way as in the Vertebrate, for the groove is never converted into a canal.

In the Ascidian we observe as the first stage in the development of the nervous system the formation of a neural groove, with its boundaries of thickened ectoderm—so far, as in the Earth-worm; but the development proceeds a stage further. The groove becomes gradually roofed in from behind forwards, forming a tube which long remains open in front and is traceable behind, through the protostome, into continuity with the alimentary cavity. But although the anterior extremity becomes enlarged, nevertheless the development of the nervous system in Phallusia turns aside from the Vertebrate road, and passes through a series of transformations which are special to the Tunicate type. Indeed these are of such a nature that the tubular character of the nervous system soon becomes no longer recognisable.

In Amphioxus exactly the same early stages are passed through; there is first the groove, then the enclosure of this to form the neural canal, open at first in front, and communicating with the alimentary cavity behind. But these apertures before long become closed, and the neural canal shut off as a distinct simple tube. In this condition it remains permanently, only that the walls of the tube become much thickened, so that the cavity is almost obliterated. Although there is no very apparent departure from the path which we have seen that the nervous system of the Toad takes in its development, nevertheless there is no further progression; the anterior enlargement to form the brain never appearing.

This brief sketch is sufficient to show that the various stages in the development of the nervous system of a Vertebrate are

represented by transitory or even by permanent conditions of the
nervous system of a series of animals lower in the scale of organi-
sation.   If we were to analyse in the same way the development
of any other specialised segregation of one of the higher animals,
we should find that exactly similar results would be arrived at.
Did time and space allow, it would be easy to trace the corre-
spondences of development in the case of the heart, of the central
axis or notochord, of the branchial slits, of the renal organs, and
so on.   And if this correspondence of permanent conditions in
lower animals, with stages of development in higher animals,
is thus found to obtain to minute detail in the separate
and specialised parts, it stands to reason that it must be
found also in the aggregate which those parts compose.   And
indeed, in many cases, even in the mere matter of external form,
the correspondence is often such as to strike the most ordinary
observer.   Compare, for example, the polyp stage of development
of the jelly-fish with the permanent conditions of some of the
Hydroid polyps ; compare the Scyphistoma stage of Aurelia with
the permanent condition of Lucernaria ; compare the tailed larva
of Phallusia with the permanent condition of Appendicularia ;
compare the several stages of transformation of one of the higher
Crustacea with permanent conditions of the lower Crustacea ;
compare the various stages of the developing Amphibian with
either transitory or permanent conditions of Worms, of Tunicates,
of Amphioxus, and of lower Vertebrates !

In conclusion I will attempt to formulate as briefly as pos-
sible, some of the general results at which we are able to arrive
from a consideration of the facts we have had before us :

I. If we compare the processes of development of any two
animals, from Sponges upwards, we find complete correspondence
up to a certain point ; from which point they may diverge from
one another.   This point is sometimes placed near the bottom of
the development-scale, sometimes near the top ; or, it may be, in
any intermediate position.

II. Development is essentially localisation of function and
concomitant or consequent modification of structure ; such modi-
fication being accompanied by segregation of the cells concerned
with the function localised.

III. The path of development of all the more important of
these segregated parts is the same up to a certain point in the
development of each segregation.   From this point it may, in any
animals or group of animals, diverge from the rest ; or may remain
stationary, whilst in the others, specialisation and modification
progress further.

IV. The various stages or phases of development of an animal,
as well as of its specialised parts, are often found to correspond

with either permanent or transient conditions of animals lower in the scale.

V. Since the phases of development of individual animals are often seen to be representations of the permanent conditions which are met with in a series of animals belonging to lower grades of organisation, it is impossible not to infer that these successive phases in the development of the individual represent similar phases in the process of formation or development of the race to which the individual belongs. To revert to a former simile, we may safely say that the developmental telescope of the individual is the same as that of the race, but with the tubes shortened or shifted one upon another so that in many cases their original order is no longer recognisable. The history of the development then of any individual animal from the egg it an abridgment of the history of formation in time of the race ; or, to state the matter in as few words as possible, "developmens represents descent."

We conclude, therefore, that the ancestors of every animal have successively exhibited structural conditions which are represented in a more or less modified form by the successive stages of development of the individual. This is the only logical conclusion to which the study of animal development leads. Modifying slightly the words of Darwin I would say, " to take any other view is to admit that the structure of animals, and the history of their development, form a mere snare laid to entrap our judgment."

On the HISTOLOGY *of* HYDRA FUSCA.[1]   By T. JEFFERY PARKER,
B.Sc., Professor of Natural History in the University of
Otago.

THE few observations I have to offer on this much-discussed
subject are partly confirmatory of, partly supplementary to, those
of Kleinenberg;[2] they present a certain agreement with those of
F. E. Schulze;[3] while they are, in great measure, distinctly con-
tradictory of the later researches of Korotneff.[4]

1. *The Ectoderm and the Muscular Layer.*—The layer of
longitudinal fibres between the ectoderm and the endoderm was
discovered by Kölliker, who believed that each fibre was in direct
connection with an endoderm cell.   Kleinenberg, in teased spe-
cimens, saw that the ectoderm cells tapered towards their inner
ends, and that each was continued into a simple or branched
process, of precisely the same character as the fibres seen in sec-
tions : from this observation the important conclusion was arrived
at, that the fibres were in direct continuity with the ectoderm
cells, thus forming a sort of nascent mesoderm.

Schulze figures the elements of the middle layer as fusiform
fibres with somewhat jagged edges. Korotneff, following Kleinen-
berg's directions as to methods of preparation, came to the con-
clusion that the ectoderm cells were expanded (*élargie*) at their
inner ends, and that each carried a fusiform refringent fibre,
attached by its middle to the enlarged base of the cell, and pro-
jecting beyond it in either direction, so that the cell appeared as
a lateral appendage (*annexe*) of the fibre, rather than the fibre as
a prolongation of the cell.

How M. Korotneff can have come to this conclusion as to the
shape of the ectoderm cells it is rather difficult to imagine; by
any ordinary method of preparation it is perfectly easy to satisfy
oneself that the ectoderm cells of the body are, as a rule, markedly
distinguished from those of the endoderm by the tapering of
their inner ends ; and, in good specimens, that these ends are
continued into longer or shorter filaments.

The question of the exact relations of the fibres is by no means
so easy to decide.   Any one working at *Hydra* for a week or two,
and using various methods of preparation, might readily frame a

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[1] From the 'Proceedings of the Royal Society,' No. 200, 1880.   The
Plate there given is not reproduced.

[2] 'Hydra,' 1872.

[3] 'Ueber den Bau u. die Entwicklung von Cordylophora lucustris,'
1871.

[4] " Histologie de l'Hydre et de la Lucernaire," 'Arch. de Zool. exp.,
t. v (1876), p. 369.

dozen different theories on this point, all equally supported by appearances. But the matter seems to me to be entirely set at rest by thin longitudinal sections of specimens preserved in ammonic bichromate, which reagent usually has the effect of causing a certain amount of separation between the layers. In such sections the ectoderm cells are distinctly seen to taper off towards their inner ends; the fibres to pass from them, at a sharp angle, towards the endoderm, or, more correctly, towards the supporting lamella; and, in some cases, the fibres can be distinctly traced into the attenuated extremities of the cells.

As to the true nature and functions of these structures, Dr. Kleinenberg calls the ectoderm cell, with its filamentous process, a neuro-muscle cell; M. Korotneff prefers to name it an epithelio-muscle cell; Professor Huxley[1] considers that the fibres "are solely internuncial in function, and, therefore, the primary form of nerves." This last view is rendered, to say the least, decidedly improbable, by the great number and the regular disposition of the fibres. It seems à priori unlikely that an animal devoid of all muscular tissue should have a layer of close-set longitudinal nerve-fibres throughout its whole body, while such an arrangement is perfectly intelligible in a set of specially contractile filaments, developed as a means of rapid retraction of the body.

The term "neuro-muscular" implies, as Kleinenberg explains, that the process only is contractile, the function of the cell itself being merely to receive and transmit impressions. But, as Professor Huxley points out, it is absolutely necessary to assume contractility in the cell proper to account for the lengthening of the body. The fibres merely have a special degree of contractility assigned to them, in correspondence with the obvious advantage accruing to the animal from the power of instantaneous shortening, the general contractility of the cells serving for extension, this movement being, as observation of a living *Hydra* shows, a comparatively slow one. The fibres must also be of use in the characteristic "looping" movements of the animal.

The simplest and most reasonable way of looking at these structures is that adopted by Dr. Michael Foster, and illustrated in the diagram at the beginning of the third chapter of his 'Text-book of Physiology.' These show clearly enough that the ectoderm cell of *Hydra*, with its muscular process, is the equivalent of what, in the higher animals, becomes sensory cell, sensory nerve, nerve cell, motor nerve, and muscle cell. So that a fairly logical term might be made by combining Kleinenberg's and Korotneff's, and speaking of epithelioneuro-muscle cell; but, fortunately, it is unnecessary to employ any such cumbersome

[1] 'Anat. of Invert. Animals,' p. 64.

term, and quite sufficient to speak of ectoderm cell with contractile process.

The interstitial tissue, discovered by Kleinenburg, is quite readily made out in all parts of the body except the proximal end, where nematocysts are also absent. It is not mentioned by Korotneff, and, indeed, its existence would be impossible if the large ectoderm cells had the shape described by him.

I have found no interstitial cells in the tentacles; this would seem to show that the ordinary ectoderm cells may also be the mother cells of the nematocysts. The ectoderm cells of the tentacles also differ from those of the body from the fact that their nuclei are non-nucleolate, resembling, indeed, the nucleoli of the body-cells, rather than their nuclei.

2. *The Supporting Lamella.*—This structure is clearly distinguished by Schulze and by Korotneff, the latter of whom, however, figures it[1] as almost equal in thickness to the diameter of an ectoderm cell! Kleinenberg states that the muscular processes are imbedded in a structureless cementing substance, and that this, continued beyond the muscular layer on the endoderm side, forms a layer—the " Stützlamelle" of Reichert—which can sometimes be obtained as a separate structure.

This description by no means expresses the distinctness of the supporting lamella. In specimens preserved in osmic acid or ammonium bichromate, without subsequent treatment with alcohol, it is easy, by teasing with fine needles, to detach shreds of considerable extent, more or less free from attached muscular fibrils and from cells of the interstitial tissue.

3. *The Endoderm.*—The ciliation of the endoderm is a question about which there has been a good deal of discussion. Schluze figures a single flagellum to each cell, as seen in optical section of the tentacle. Kleinenberg was unable to demonstrate the existence of flagella in the uninjured animal, or in preserved specimens, but in transverse sections of the living animal he observed one or two cilia in connection with more or fewer of the cells, and noticed that they were not fixed structures, but were occasionally retracted, and then protruded again, the cells at the same time sending out pseudopodial processes.

It is quite easy to confirm this observation; the slow lashing movement of the flagelliform cilia their continual disappearance and reappearance in fresh places can be made out without difficulty. But the best notion of the characters and relation of the cilia is obtained by teasing out, or, still better, by cutting thin sections of osmic acid specimens. Such preparations quite lead one to think that the endoderm is ciliated throughout; in the sections particularly cell after cell is seen bearing one, two, or

[1] Loc. cit., pl. 15, fig. 8.

three cilia. These latter are of great length, in fact, nearly or quite as long as the cells to which they are attached; in some cases, indeed, they are longer. I have never seen anything like a "collar" at the base of any of the cilia.

The amœboid character of the endoderm cells, as seen in sections or teased fragments of the living animal, is a well-known fact; but the extent and activity of the amœboid movements during life has not been sufficiently insisted on. In sections of picric acid or ammonic bichromate specimens, large rounded pseudopodia are seen to be given off from the cells into the digestive cavity, sometimes to such an extent as completely to obliterate the latter. The length of the cells may, therefore, vary almost indefinitely; they may be but little longer than the ectoderm cells, or may be two or three times as long. This variation in the size of the endoderm cells, and the consequent variation in the diameter of the digestive cavity, is very marked in my series of sections, nearly all of which are taken from large specimens,[1] killed in a state of half extension. When the endoderm cells are fully extended, it is almost impossible to obtain them complete by teasing. They nearly always break across, and can only be obtained in a fragmentary condition.

A very noticeable point about the endoderm cells is the presence in their protoplasm, especially towards the far end, of dark-coloured irregular granules, of various sizes. It has been suggested that these are products of excretion : Kleinenberg makes the important observation that their number varies with the state of nutrition of the animal.

I am convinced that these bodies are food particles, taken into the protoplasm of the cells, from the partially disintegrated bodies of the *Entomostraca* in the digestive cavity. They are of quite the same nature as the contents of the alimentary canal in many of the common *Cladocera* and *Copepoda*; they occur chiefly in the free end of the cell, and in some cases they have all the appearance of being half in and half out of the protoplasm. The particles of the more transparent parts of the body of the Crustaceans will naturally not be so evident in the cell protoplasm; even these, however, can be made out in a *Hydra* in full digestion, when the endoderm cells of the distal or gastric region are completely crammed with transparent spheroids.

The clearest case of ingestion of solid particles was seen when a diatom was seen to be completely imbedded in the protoplasm of a cell.

If this explanation of the dark granules is the correct one *Hydra* will have been shown to exhibit a process of alimentation identical with that described by Metschinkoff in the lower *Tur-*

[1] Supplied by Mr. Bolton, of Birmingham.

bellaria and in sponges.[1] The Russian observer describes the complete obliteration, during digestion, of the digestive cavity in the Turbellarians, and of the canals in the sponges; and, in the former as well as the latter, he has undoubted evidence of the actual ingestion of solid particles by the endoderm cells.

It would seem, therefore, that *Hydra* adds another instance to the two already brought forward by Metschinkoff, of a Metazoon exhibiting what is usually considered to be a distinctively Protozoan mode of digestion. It is quite possible that a preliminary disintegration of the animals taken in is performed by juices secreted by the endoderm cells, but the final digestion seems to take place in the actual protoplasm of the cells, into which the food articles are taken in the solid form.

The endoderm cells of the tentacles resemble those of the proximal end of the body in possessing large vacuoles. Their nuclei are in some instances, although not constantly, simple and non-nucleolate like those of the ectoderm cells from the same region.

Finally, I have been able fully to confirm Professor Huxley's statement[2] as to the presence of nematocysts in the endoderm (fig. 1, *n*), a statement which, as far as I am aware, has not been made, with regard to *Hydra*, by any other writer on the subject. This fact is, like the absence of interstitial tissue in the tentacles, an argument against Kleinenberg's view that the tissue is the sole source of the nematocysts.

4. *Methods.*—For sections, the *Hydræ* were either killed with hot water, and placed in Kleinenberg's picric acid for two hours, or were placed alive in ammonic bichromate, 1 per cent.—which always kills them in the half extended condition—and kept in it for two or three days. In either case they were afterwards transferred to 50 per cent. alcohol, and then placed successively in 75 per cent., 90 per cent., and absolute alcohol. The specimens were stained either with carmine or picrocarmine, and imbedded in cacao butter, after soaking for a short time first in oil of cloves and then in melted cacao butter. By this means they became so thoroughly permeated with the imbedding material that they could be cut without the loss of a single section; even longitudinal sections of the tentacles could be made with ease.

For teasing I employed ammonic bichromate, acetic acid (0·5

[1] 'Zool. Anzeiger,' Bd. I, (1878), p. 387, and 'Zeitsch. f. wiss. Zool.,' Bd. xxxii (1879). It need hardly be said that the above view of the physiology of digestion in *Hydra* was suggested by these papers of Metschinkoff's.

[2] Huxley and Martin, 'Elementary Biology,' p. 100,

per cent.), or osmic acid (1 per cent.), and for this purpose the specimens were not transferred to alcohol, but to weak glycerine (equal parts of glycerine and water), in which they were teased out.

For sections showing the cilia of the endoderm, the *Hydra* were kept for twenty-four hours in 1 per cent. osmic acid, then washed, and preserved in weak glycerine until required for cutting. The sections were cut by Dr. Pritchard's very convenient freezing microtome, the specimens being placed in gum water before freezing.

The ORTHONECTIDA, *a* NEW CLASS *of the* PHYLUM *of the* WORMS.[1] By ALFRED GIARD, Professor in the Faculty of Sciences of Lille. With Plate XXII.

IN 1868, in an interesting memoir[2] on the marine Planarians of St. Malo, Keferstein figures an animal which he designates by the name " problematic parasite." The explanation of the plate (plate ii, fig. 8) indicates that this parasite has been found several times, and often in great quantity, in the digestive tube of the Tremellaria (*Leptoplana tremellaris*). The text gives us no information as to this curious organism, and it is chiefly by means of the rather rough figure of the second plate of Kerferstein's work that we are led to refer it to the group which we are about to study.

MacIntosh, in his beautiful monograph of the British Nemerteans, published in 1874,[3] recalls the observation of Keferstein in reference to a parasite which he met with in *Lineus gesserensis* (*Nemertes communis* of P. J. Van Beneden), and concerning which he gives the following details, accompanied by a few drawings :

" Another curious parasite is found burrowing in the bodywall of *Lineus gesserensis*, its presence being readily recognised by the perforated and honeycombed appearance of the dorsum of the affected animal, whose textures seem to be the seat of the workings of a microscopic *Tomicus typographicus*. When highly magnified, the affected region appears to be covered with a vast network of pale, minutely granular channels, which contain numerous opaque, ovoid, granular masses.

" On rupturing the body of the worm a large number of the peculiar structures (plate xviii, fig. 17) slide out of the channels, and swim through the surrounding water, generally, though not always, with the upper end (in the figure) first. Externally they are coated with long cilia, whose activity in the free state is of somewhat short duration, for after a time the animals remain quiet, and they drop off. The body is

[1] This memoir has been corrected and new matter, including previously unpublished figures, has been added by Professor Giard to the original memoir, which appeared in ' Robin and Ponchet's Journal,' for the present publication.

[2] Keferstein, ' Beiträge zur Anatomie und Entwickel. einiger Seeplanarien,' Göttingen, 1868.

[3] ' A Monograph of the British Annelids,' part i, Ray Society, p. 129, and pl. xviii, figs. 17, 18, 19.

distinctly segmented, and tapers slightly towards the posterior end ; while the surface is marked by very fine longitudinal striæ, as in Opalina, though in a much more minute degree. Anteriorly is a conical portion (*a*), composed of three rather indistinctly marked segments. Two evident annuli (*b*) succeed, the posterior part of the last being narrowed, so as to cause a constriction of the body-wall. Behind are six nearly equal divisions (*c*), each often appearing double, that is, has a broad anterior and a narrow posterior annulus. The posterior region (*d*) consists of three indistinct segments. The body is minutely granular throughout, and an internal cavity is apparent from the fourth segment to the last, commencing in the former by a rounded end, and terminating just within the border of the latter. No aperture is observed at either end. The opaque, ovoid, granular bodies (plate xviii, fig. 18), scattered profusely throughout the infected portions of the Lineus, are evidently early stages in the development of this species, and they, too, are ciliated. On subjecting them to gentle pressure (fig. 19), transverse segmentation is apparent, the number of segments varying according to the degree of advancement. The parasites are very delicate structures, and in the free state soon break up into cells and granules, after discarding their cilia, as above mentioned. Transverse section of the affected worms shows that they occur both in the skin and in the walls of the digestive tract, their ravages in the pigmentary layer of the former tissue causing the curious appearances which led to their detection. It is a somewhat difficult point to determine whether the skin, muscles of the body-wall, or digestive canal, constitute the common area of this creature's depredations, whether it is piercing the former on its way to the surface, or passing to the alimentary cavity to be voided per anum. The characteristically segmented condition of the mature specimens and their internal structure exhibit a higher type of organisation than the ordinary Opalina. Professor Keferstein found a very similar parasite in the stomach of *Leptoplana tremellaris*, but he did not describe it further than simply mention, under the explanation of the plate, that it is an enigmatical structure. The centre of the body is occupied by a double row of large cells in his figure."

Such are the details, certainly rather incomplete, it must be conceded, which we have been able to find in earlier writers touching the curious animals which we have designated by the name of ORTHONECTIDA.[1]

[1] We published in the ' Comptus Rendus de l'Académie des Sciences '

II.—Species Observed.

I have had the opportunity of studying the species discovered by MacIntosh. *Lineus gesserensis* is very common at Wimereux, as also its variety *Lineus sanguineus,* under stones, in oozy spots, which are accessible every day even during the neap tides.

My attention was drawn to the parasite of this species, a rare one, on the whole, by my pupil and friend, J. Barrois, at the time when he was preparing in our laboratory his work on the Nemerteans. But I could then, in the absence of sufficient material, do no more than verify a part of the observations of MacIntosh, and rectify certain errors committed by this eminent zoologist, as I shall point out further on.

In vain I have sought, on several occasions, for the parasite discovered by Keferstein in *Leptoplana tremellaris ;* and yet this Planarian is excessively common at Wimereux, in very nearly the same spots as *Lineus gesserensis.* Paul Hallez, Demonstrator in the Faculty of Sciences at Lille, who has dissected numerous examples of Leptoplana, has had no better luck than I in this search.

A happy chance made me acquainted, in 1877, with two new species of Orthonectida, which I have been able since to obtain in sufficient abundance to make a nearly complete study of them, although some gaps still remain to be filled up.

During the autumn of 1877 I had betaken myself to Wimereux in order to study the embryogeny of a species of Ophiura with condensed development. I had a choice between two species, *Ophiothrix fragilis* and *Ophiocoma neglecta,* both equally common, hermaphrodite, and viviparous. Reasons of an entirely technical nature caused me to prefer the second. In opening hundreds of Ophiocoma, to extract from them the embryos, I discovered two species of ORTHONECTIDA which have enabled me to undertake the study of the group. Both are excessively abundant in the animal infested by them, but it is quite a rare thing to find an Ophiurid thus infested. According to my notes, each species may be found once among about eighty specimens of Ophiurids, so that one has the chance of finding one or other species once in forty specimens dissected.[1]

(Seance du 29, Octobre, 1870), a short preliminary notice of these animals.

[1] Besides these two species of Orthonectida, *Ophiocoma neglecta* presents at Wimereux a certain number of other parasites, which are interesting.

1. A pretty Vorticella with a very short peduncle (*Vorticella ophiocomæ,*

In the month of June last I twice found the two species of Orthonectida associated in the same specimen of an Ophiurid.

It has appeared to me as though the Orthonectida were more common in the autumn than at any other season. But this perhaps depends on the fact that I had been able to give more attention then to my researches than during the academical session.

III.—ANATOMY OF RHOPALURA OPHIOCOMÆ.

Nothing is easier than, with aid of fine needles, to separate the whole dorsal region of Ophiocomma from the ventral disc formed by the buccal plates and the arms. The animal deprived of its dorsal cupola, and consequently of its digestive and reproductive organs, lives nevertheless several days, walks, and conducts itself generally as though no injury had occurred to it.

In tearing open the dorsal cupola, in order to force out the embryos, the glass slip on which the operation is being conducted is sometimes seen to be covered by small white flakes, which, on examination with a low power of the microscope, are seen to be animated by a rapid movement of translation in a straight line, which is highly characteristic.

This movement, common to all the species of this group, has led me to give to these organisms the name of Orthonectida. These agile little animals resemble large ciliated infusoria of a porcellanous aspect and a generally cylindrical form. A certain number, however, have a somewhat shorter form, irregularly ovoid, and move more slowly.

In the one form or the other they are simple *Planulæ*,

nov. sp.), which is found even on the arms of young individuals still enclosed in the maternal incubatory pouch (very common).

2. An Urceolaria (*Ur. ophiocomæ*, nov. sp.), also very common.

3. A copepod crustacean of the family of the Ergasillidæ, or rather of a distinct family comprising the annelidocolous species. This crustacean, which is broad and provided with hooked limbs, attaches itself to the arm of the Ophiurid between the bristles. It recalls the form of the crab-louse. By a very natural association, I name it *Phthiriopsis Emilii*, and I dedicate this parasite of the star-fishes to M. Emile Blanchard, who teaches at the Museum of Paris the history of the Arthropoda. The Phthiriopsis is very rare.

4. A curious little annelid of the genus Sphærodorum, which presents the same relations to Ophiocoma which *Chlorhema Dujardinii* does to *Psammechinus miliaris*. I shall call it *Sphærodorum Greefii*, dedicating it to the learned Professor of the University of Marburg. About one Ophiurid in fifteen harbours a *Sphærodorum*.

All these parasites of Ophiocoma will be studied in detail in a special memoir.

that is to say, organisms composed fundamentally by two
layers of cells, the one within the other—an ectoderm or
external layer of cells, which are mostly ciliated, and an
endoderm constituted by larger cells, which are more granular,
and form the lining of a central cavity devoid of either mouth
or anus.

The first ring terminates in a blunt cone anteriorly, which
carries a bunch of stiff cilia. It is followed by a cylin-
drical ring of the same length, the whole surface of which is
beset with papillæ, which are disposed in eight or ten longi-
tudinal lines and in four or five transverse series ; this ring
is the sole part of the body which does not present vibratile
cilia.

The third ring is larger by itself than the two first
together ; it gradually widens towards its posterior ex-
tremity.

The fourth metamere is of the same dimensions as the
papilliferous ring ; it is followed by a terminal ring provided
with longer cilia, forming a brush at its posterior extremity.
This last ring is conical and subdivided into two metameres,
less distinct than those in front.

The last rings form a sort of club, with which the animal
strikes the water, independently of the movement of the
cilia, giving sharp strokes, which one is immediately led to
attribute to the contraction of muscular elements. We
shall point out below where these elements occur.

Such is the elongated variety. The ovoid variety differs
from it only by being a third less in length and a greater
breadth. It seems, at first sight, as though it had been
derived from the former by a contraction along the long
axis ; but it is easy to assure one's self that this is not the
case, and that it is, on the contrary, the elongated form
which is the ultimate condition of the ovoid form, which
latter is only the final embryonic phase of the animal.

For a more complete study of the anatomy of Rhopalura
it is necessary to employ very high magnifying power, the
objective 6 of Verick, or better, the objectif 9 (immersion) of
Hartnack.

The ectodermal cells then appear very distinctly, except
in the papilliferous zone where it is very difficult to deter-
mine their number and form. All the other metameres are
made up by a single transverse row of cells, and the differ-
ence in the length of the metameres depends entirely on the
difference in the length of the cellular elements which com-
pose it. The terminal rings are formed entirely by four
cells, as in the Dicyemida, the median rings comprise six or

eight cells; it is very difficult to count with exactitude, since optical sections rarely present themselves, and it is impossible to make real sections.

The ectodermal cells present very long and very dense cilia. By using osmic acid, followed by picrocarmine, it is easy to preserve the ectoderm with its clothing of cilia. Preparations made a year ago give at this day an excellent idea of the living animal.[1]

The endoderm is primitively formed of larger cells than those of the ectoderm, but they undergo in the adult a very singular modification. The whole of this layer forms an oval sac, the anterior extremity of which is hidden by the papilliferous ring, and extends from the penultimate metamere until it is inserted, in the form of a sort of pedicle, between the four terminal ectodermal cells (Plate XXII, figs. 3 and 4). The swollen part of the endodermal sac presents, when examined with the immersion lens, fine muscular bands disposed in a finely granular matrix, and recalling the appearance of the endodermic muscular layer of certain nematoids.

I do not believe that these muscular elements are derived from the bodily transformation of certain endodermal cells; I consider them rather as a *part* only of such cells, which are thus called upon to play a double physiological *rôle*. They would thus be analogous to the epithelio-muscular cells described by Korotneff in Hydra, but with this difference, that in the present case it is the external part of the cell which becomes muscular, the internal part remaining epithelial.

I would draw the attention of histologists, more skilled than I am in technical methods, to this very delicate point. The question involved is one worthy of their skill.

Metschnikoff has recently put forward the opinion that the striated bands are formed by the tails of spermatozoa, the entire endoderm being nothing but a testicular gland.[2]

I am quite unable at present to accept the opinion of my learned opponent. These bands are chiefly visible in young individuals; their number is constant; they are always disposed obliquely, as I have figured them, on an ovoid endoderm; and this disposition is not, as I had at first supposed, the result of an accidental torsion. On changing the point of view, the continuation of the spiral is seen

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[1] I have shown these preparations to various persons, and specially to Dr. Macleod, of the University of Ghent, who spent a few days at Wimereux during last April.

[2] 'Zoologischen Anzeiger,' II, No. 43, p. 619.

on the other side of the body, and the clear interspaces take the form of lozenges, just as one sees sometimes in looking through certain kinds of open basket-work. Finally, it will be seen below, that I have also found these elements in *Intoshia*, which Metschinkoff considers as the female form of Rhopalura, and where, consequently, there would be no spermatozoa.

Whatever may be the origin of these muscular elements, they unite, as I have pointed out above, into a sort of fascicle at the terminal part of the adult animals, and by their contraction give rise to those brusque movements, those strokes of the club-like tail, to which the name *Rhopalura* has reference.

If the interpretation above given is admitted, there would not be in the Orthonectida any true middle layer, but only a splanchno-pleural pseudo-mesoderm, comparable to the somato-pleural pseudo-mesoderm of the Hydra.

I give to the totality of these elements the name o *pseudo-mesoderm*, because it appears to me desirable to reserve the name of mesoderm, properly so called, for other structures which do not exist in the Orthonectida, and the homology of which in the various groups of Metazoa is very difficult to establish.

I distinguish :—(1) a *solid mesoderm*, formed very early at the expense of the endodermic cells of the embryo (rudiment of the notochord of the Tunicates and of the Vertebrates, skeletogenous cells of the embryo of Echinoderms, mesodermal cells derived from the four first spheres of the endoderm of Planarians, of Bonellia, according to the researches of P. Hallez and of Spengel, &c.). (2) A *cavitary mesoderm* formed by the diverticula of the endoderm (enterocœls), and appearing generally at a later epoch (aquiferous system of Echinoderms, enterocœl of the Tunicates, of the Brachiopods, of Sagitta, of Amphioxus, &c.). The solid mesoderm gives rise chiefly to the muscular system ; the cavitary mesoderm forms principally the vascular organs.

The physiological *rôle* of a histological element has, be it noted, only a secondary importance for the determination of phylogenetic homologies. A muscular element, for example, will always be formed at that point where it is needed, sometimes in a rudiment having an endodermal origin, sometimes at the expense of ectodermic elements (Nemerteans). It may even be formed by a mere portion of a cell (plastidule), as we find in the Infusoria, in the Cœlentera, and in the Orthonectida.

The interior of the endodermic sac is filled, in certain

individuals, by cellular elements, which give rise to the genital products. I am at a loss to understand why Metschnikoff refuses the name of endoderm to a cell-layer which arises absolutely in the same way and plays precisely the same part as the part called by this name by all embryologists in the other Metazoa.

I have seen in some Rhopaluræ a cloud of agile corpuscles issue from the sides of the body between the third and the fourth metameres. Are these corpuscles spermatozoa? It was not possible for me to get a distinct notion of their form, and I also am unable to affirm the presence of a natural opening at this spot on the body. It is possible that the specimens which presented this appearance of an emission of spermatozoa were really the victims of lacerations.

IV.—ANATOMY OF INTOSHIA GIGAS.

The second species of Orthonectida parasitic in *Ophiocoma neglecta* is much larger than *Rhophalura*. Its length is, in fact, two and a half times that of the latter. I name it *Intoshia gigas*.

Intoshia presents in swimming alternative movements of contraction and expansion in a transverse direction. It does not possess a papilliferous ring, but simply a ring devoid of cilia in its place. The body is of a more regular breadth and less tapering at the two extremities, which are blunt points, a little more conical than in the *Intoshia* parasitic in Nemerteans and Planarians.

Further, the anterior part of the body is strongly flattened in *Intoshia gigas*, and the non-ciliated segment presents on its inferior face, throughout its breadth, a transverse groove of some depth, so that the profile of the animal is that of a shoe with a heel to it (Plate XXII, fig. 5).

The metamerisation is less distinct than in Rhopalura. After the cephalic ring and a cervical ring corresponding to the papilliferous ring there follow three metameres of decreasing breadth (the third (γ) is about half of the first (a), the second (β) being of intermediate dimensions). To these follows a ring (δ) of much larger size, which seems sometimes to be divided into three, then follow two very small metameres (ϵ and ζ), which have about the same length as γ, and, finally, the terminal piece.

The variable dimensions of these metameres is no longer related, as in *Rhopalura*, to the size of the compound cells.

Each metamere is, in fact, formed by several rows of cells, the cells of each row being regularly placed over the corre-

sponding cell of adjacent rows, so as to form a longitudinal series. It is to this disposition that the longitudinal striæ observed by MacIntosh are due, which led this naturalist to approximate the parasites of Lineus to the Opalinæ.

The ring *a* contains three rows of cells, the ring β comprises two, the ring γ only one, and the others only one.

The ectodermal cells of *Intoshia gigas* are, accordingly, much smaller and much more numerous than those of *Rhopalura*. All the cells, without exception, have long cilia. The head carries, as in Rhopalura, a bunch of straight cilia directed forwards. This character is, in fact, common to all known species of Orthonectida.

The endoderm forms a regularly ovoid sac, constituted in the adult animal by beautiful polygonal cells, and enclosing, in its interior, other cells of a rounded shape, more or less abundant.

The action of acetic acid is such as to detach the ectoderm (Pl. XXII, fig. 7), and then brings well to view the endodermic sac. The jerking movements of the caudal part are much less energetic in *Intoshia* than in *Rhopalura*. We should, therefore, expect to find the muscular system much reduced; and this is actually the case, and for a long time I in vain endeavoured to see the muscular bands at all. I succeeded, however, last summer by producing a gradual and very slight compression of the animal by allowing the water to evaporate from beneath the cover glass. The bands were rendered visible at the anterior part of the body, where the endodermic cells hinder observation the least. The bands have the same oblique direction as in Rhopalura.

V.—Gemmiparous Reproduction of the Orthonectida.

The oldest individuals of *Rhopalura* or of *Intoshia* present a considerable modification of the endodermal cell-layer. The cells of this layer are no longer visible, and the endoderm seems formed by a homogeneous membrane of granular aspect, very similar to certain tissues of the Nematoids. On the other hand, we have seen that the ectoderm can be detached in these animals with the greatest facility under the influence of reagents. As a result of the proliferation of cellular elements in the interior of the endodermal sac, this organ swells, becomes spherical, and bursting the ectoderm, which disappears, is transformed into a true " sporo-cyst " (Pl. XXII, figs. 14, 15, 16 and 17). In the interior of the sporocyst are seen buds, the cellular nature of which is extremely difficult to demonstrate. The same

difficulty, it may be noted, is found with regard to the buds of the sporocysts of the Trematoda, and all zoologists who have occupied themselves with the study of these animals know how difficult it is to demonstrate the cells which form the bud-like embryo of a *Bucephalus* or of any Distoma.

Occasionally the primary buds produce secondary buds. When these buds have arrived at a certain size, they are composed clearly enough by a single layer of cells, which then proceed later to form an internal layer by delamination.

It appears, from what has been just stated, that the endoderm alone of the parent animal enters into the formation of the bud-like embryos, but it must not be forgotten that the papilliferous ring, or that which represents it in *Intoshia*, is characterised by great opacity, and that it is possible that at certain points ectodermal cells penetrate to the interior of the endodermal sac.

With their increasing development, the sporocysts lose their original form and become very voluminous. Often there are found several sporocysts in the interior of a specimen of an Ophiurid. This very active gemmiparous reproduction of the Orthonectida explains how it is that these animals are found in such great abundance in the Echinoderms infested by them. The phenomenon is one very similar to that occurring in the case of the Dicyemida; but with regard to these latter, it is almost impossible to find a Cephalopod which does not swarm with them, whilst the Orthonectida are comparatively rare.

VI.—Oviparous Reproduction.

Side by side with the sporocysts just described, we find in the infested Ophiurids a great number of cellular masses, which must be considered as eggs in various stages of segmentation.

In the case of *Rhopalura*, I have only been able to observe a small number of these embryonic stages (Pl. XXII, figs. 19, 20, 21). The little which I have been able to observe leads me to the conclusion that the segmentation is irregular, and that the planula is formed by epibolé, as in the Dicyemida. Each division of the cells in the mulberry mass gives rise to stellate figures, so well known at the present day by the name of *caryolitic figures* or *amphiasters*. The use of osmic acid enables one to observe them with the greatest facility.

In the study of the development of *Intoshia* I have been more successful. Here the segmented egg forms at first a perfectly regular *blastula*, the cells of which are at first

very much elongated in a radial direction (Pl. XXII, figs. 8 to 13). This *blastula* very soon exhibits a very definite process of delamination; the internal part of each cylindrical cell gives rise to a spherical cellular mass, which detaches itself from the mother-cell, which is shortened by the process (Pl. XXII, fig. 10). This is absolutely the same mode of formation as that which can be observed in the production of the mesoderm in the Ophiurids with a condensed embryogeny (*Ophiothrix fragilis, Ophiocoma neglecta*). In the present case it is the endoderm which forms at the expense of the ectoderm, but we have seen that this endoderm contains potentially the mesodermic elements (the muscular bands).

Soon the ectodermal cells become covered with vibratile cilia, and elongate in a definite direction; then the embryo, which was spherical, becomes ovoid, the metameres show themselves little by little, and by insensible gradations we arrive at the adult condition.

VII.—Systematic.

According to what has been stated in the preceding pages, the Orthonectida may be defined in the following manner :—
" Metazoa retaining throughout their existence the form of the *planula*, having a ciliated ectoderm (stiff cilia forming a bunch on the anterior cephalic region, vibratile cilia on the other regions of the body), presenting metameres, which do not correspond to any internal division of the body, with a sacciform endoderm, which gives rise to a muscular splanchno-pleural pseudo-mesoderm. Reproduction of two kinds: (1) gemmiparous in the interior of sporocysts, constituted by the development of the endoderm ; (2) oviparous, dependent on the combination of male and female elements, formed in all probability in different individuals."

The class comprises at present two genera :

I. The genus Rhopalura, characterised by the presence of a papilliferous ring, by its ectoderm composed of large cells limited in number, and its definitely muscular endoderm.

Species unique. *Rhopalura ophiocomæ*. Length 0·108 mm.

II. The genus *Intoshia*, devoid of papilliferous ring ; ectoderm formed of small, very numerous cells.

Three species :

1. *Intoshia gigas.* — Parasitic in *Ophiocoma neglecta*. Length 0·270 mm.

2. *Intoshia linei.*—Parasitic in *Lineus gesserensis*. Length 0·157 mm.

3. *Intoshia leptoplanæ.*—Parasitic in *Leptoplana tremellaris.* Length 0·135, breadth 0·03 mm.

This last species, like the preceding, is regularly cylindrical, and rounded at the extremities. According to the figure of Keferstein, it presents ten metameres, perfectly regular, in addition to the cephalic and caudal rings. The endoderm appears to be formed by large spherical cells.

The general form of the body, the regularity of the metamerisation in the two species parasitic in the Nemertean and the Planarian, lead to the supposition that there may be other differential characters present in them, which would lead zoologists probably to create a particular genus for these two types. Such a step in our present state of knowledge would, it appears to me, be superfluous.

Metschnikoff[1] has recently described, under the name of *Rhopalura Giardii,* a new species of Orthonectida, which he has since identified with *Rhopalura ophiocomæ.*

This species, which is parasitic in *Amphiura squamata,* was found abundantly at Spezzia. It presents itself in two different forms, which Metschinkoff thinks are probably identical with *Rhopalura ophiocomæ* and *Intoshia gigas,* which, according to him, are merely the male and female of one species.

I must confess that this idea has often occurred to me during my researches, and I do not entirely give it up even at this moment. The strongest argument in its favour is that the two forms, *Intoshia* and *Rhopalura,* exist in about the same quantity in *Ophiocoma neglecta ;* and that it would be curious to find in this little Ophiurid two different representatives of a group so rare as are the Orthonectida. At the same time, the difference between the two forms is greater than I had at first supposed. Further, we have not elsewhere any example of an animal in which some females produce eggs, giving birth to males exclusively, and others eggs from which only females issue. We might, perhaps, try to remove the difficulty by supposing that, in one or the other case, there was parthenogenesis (arrenotoky or thelytoky) ; but this would be, at present, pure hypothesis. However this may be, I have no fundamental objection to such a mode of explaining the facts, but I shall wait to make up my mind for the time when I shall have found in another species of *Intoshia (Intoshia linei,* for instance) an accompanying form analogous to *Rhopalura.*

Without doubt sexual differences, such as exist in *Bonellia viridis,* and in other worms, such as *Bilharzia hæmatobia,*

[1] 'Zoologischen Anzeiger,' II, No. 40, and No. 43.

are of a nature to make one very cautious on this subject; but, in addition to the special characters of the histological constitution of the ectoderm, I have recently observed new facts which still further separate *Intoshia gigas* from *Rhopalura ophiocomæ*.

It appears, as the result of an examination of many hundreds of adult individuals, that *Intoshia gigas* never has its non-ciliated segment provided with papillæ, nor even with refringent corpuscles. The " refringent corpuscles" of M. Metschnikoff form actual projections on the ring, which I call the " papilliferous ring " in Rhopalura. It must, then, be admitted that if such corpuscles exist in the supposed females of the parasite of *Amphiura squamata*, this parasite belongs to a new species (as is probable enough *à priori*), and that the sexual dimorphism is less accentuated in this species than in the other.

VIII.—PHYLOGENY.

With reference to the position of the group of the Orthonectida on the genealogical tree of animals, it is very difficult to make a definite statement. But there is no doubt that these parasites ought to be attached to the phylum of the *Vermes*, and take their place at the base of this phylum side by side with the Dicyemida. The phylum of the worms would then, according to my idea, be represented by the following diagram :
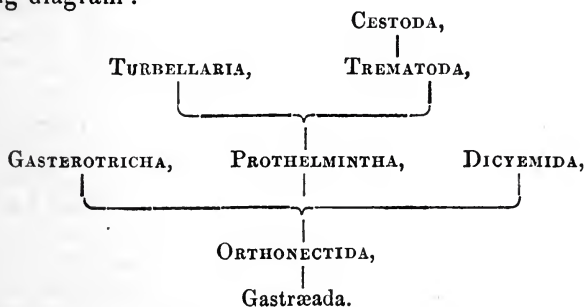
The Orthonectida must occupy in this scheme an inferior position to that of the Dicyemida. These latter are evidently much degenerated by parasitism. Their organisation must have been formerly much higher than it is to-day. The epiderm contains very clearly (*Dicyema* of *Sepia*) the rod-like bodies characteristic of the skin of the Turbellaria, and the embryo presents a very complex organ, the *urna :* nothing of this sort is seen in the Orthonectida.[1] One of the most

[1] See on " Dycyemida," the beautiful memoir of Edouard Van

characteristic features of the group of the Vermes thus limited is the existence, in all the animals of this group, of a gemmiparous reproduction (sporocysts, echinococci, &c.). This peculiarity only disappears in the higher worms, the Turbellarians, which are related to the rest of the group by so numerous and so important morphological features that no serious zoologist will entertain the idea of separating them from the Trematoda and Cestoda, in order to approximate them to the Annelids, as has sometimes been done.

Amongst the animals formerly classified with the preceding, some (Bryozoa, Annelida, and satellite-groups) are intimately related to the true Molluscs, with which I unite them to constitute the group of the *Gymnotoca*, whilst others form an assemblage which we may call the *Nematelmia*, including therein the *Nematoidea*, the *Echinorhyncha*, the *Desmoscolecida*, the *Gasterotricha*, &c. The Tunicata must be placed at the base of the phylum of Vertebrata. Budding from the interior of sporocysts is found, it is true, in other classes of the animal kingdom. Leaving aside the somewhat aberrant cases found among Arthropoda, we see in certain Rotifers (*Callidina*) an internal gemmiparity very similar to that of the Vermes. Further, the Gasterotricha, the parent-stock of the Rotifers and perhaps of all the Gymnotoka, appear to me to be readily connected with the Vermes by means of the Orthonectida.

We must not forget that the external resemblances between the Orthonectida, Gasterotricha, parasitic Rotifera, &c., are further increased by the similar manner of life obtaining in all these animals. *Ophicoma*, like *Lineus gesserensis* and *Leptoplana tremellaris*, inhabits muddy bottoms. The same is true of the limicolous Annelids and of Nebalia, on which we find as parasites certain degraded Rotifers (Balatro and Saccobdella). But the Orthonectida are in any case very inferior to the most degraded Rotifers, and represent, without any doubt, after the Gastræada, the first step of the sub-kingdom Metazoa.

I must add here that, according to a letter from Leuckart, the embryo of Distoma resembles the Orthonectida in a most astonishing manner, a fact which tends to confirm the position which I have assigned to this group among the Vermes.

Beneden, ' Bulletins de l'Acad. de Belgique,' 2e serie, t. xli and xlii, No. 7, 1876 ; also Abstract, with Plate, in ' Quart. Journ. Mic. Science,' vol. xvii, 1877, p. 132.

IX.—General Reflections.

At first sight it seems as though the discovery of the Orthonectida would bring a very solid support to the Planula theory of Ray Lankester, or, still more, to the Parenchymula theory of Metschnikoff, and I do not doubt that more than one zoologist will interpret, in that sense, the observations I have recorded.

I persist, however, for my own part, in considering the *invaginate Gastrula* as the prototype of the Metazoa. I base this opinion on the following arguments :

1. The Orthonectida are parasitic animals, and we must take into account the retrogression which this kind of life may have brought about in their structure. An organisation which we consider as one of primitive simplicity is very possibly simple only in consequence of degeneration, if we have to deal with a parasite, and especially an internal parasite.

2 We have seen that the *Planula* is formed by epibolé in Rhopalura ; in this case we have, then, at any rate, momentarily, a real gastrula, which closes up and does not reopen, because the mode of life of the animal does not require the existence of a permanent digestive tube.

3. The forms which present the embryonic phase termed *parenchymula* by El. Metschnikoff cannot be considered, as that naturalist would wish, as the lowest among the Sponges and Hydroids.

The forms known as *Halisarca* are not *low* sponges, but sponges which have undergone a degeneration of their skeletal apparatus. From the point of view of general morphology I have shown that we may compare them to the *Botrylli*, and must assign to them a very high grade among the Fibrosa, analogous to that which the Botrylli occupy among the Synascidiæ or the Leucones among the Calcispongiæ.

The Siphonophora, we may remember, are very far from being Cœlentera of inferior position, and it is by no means astonishing that they present a condensed embryogeny. The typical embryonic form is found among the Coralligena and certain Actiniæ.

The Echini and the Ophiurids with pelagic embryos have a small nutritive vitellus. They present a gastrula formed by invagination, and it is only when this gastrula is thus formed that the mesoderm takes origin, at first by partial delamination of the ectoderm, and later by lateral thickenings of the endoderm.

In the Ophiurids with condensed embryogeny, the egg presents an enormous nutritive yelk, and the mesoderm is formed by abbreviation at the same time as the endoderm by a general delamination of the ectoderm, which leads us to Metschnikoff's form dubbed *Parenchymula*. This I have established most clearly in cases of *Ophiocoma neglecta* and *Ophiothrix fragilis*.

It is clearly impossible for me to discuss here the important questions of general embryogeny which are raised by the study of the Orthonectida. I will merely say that, according to my observations, a large number of calcareous and siliceous sponges present what one may well call a *biconvex archigastrula*. Quite recently Keller has shown that *Chalinula* possesses an *amphigastrula*. Kowalevsky has described, in the most exact manner, the existence of an archigastrula in a variety of *Actinia mesembryanthemum*, in Cereanthus, and in various Medusæ. I am able to confirm his statement in reference to *Actinia equina*. Finally, Ed. van Beneden has described and figured an amphigastrula in the Dicyemida, so closely related in many respects to the Orthonectida.

These examples will suffice, I think, to justify my opinion that the *gastrula* by invagination is the primitive mode; the gastrula by delamination (*Planula* or *Parenchymula*) a secondary mode of embryonic development.

I would also direct attention to the metamerisation which is so remarkable in the *Orthonectida*.

We have seen that this metamerisation only affects the ectoderm, and I believe that this was primitively the case also in the Annelids. What proves this to be the case is the highly differentiated form of the digestive tube of the Chætopods in which metamerisation is only well marked in those organs derived from the ectoderm, such as the bristles, the parapodia, and the segmental organs (nephridia).

It is obvious enough that I am not alluding to that kind of metamerisation which is observed in *Salmacina, Syllis,* &c., which is merely the result of gemmiparous reproduction. This last kind of metamerisation is only comparable to what one finds in the Cestoidea and the Rhabdocœla.

NOTES AND MEMORANDA.

The Origin of the Red Corpuscles of Mammalian blood.—
A step forward in our knowledge of this subject has been made
by Professor Rindfleisch, of Wurzburg. As he justly remarks
(' Archiv. f. mikrosk. Anatomie,' vol. xvii, August, 1879), in
the introduction to his memoir, "not a word is needed as to the
usefulness, in fact the necessity, of continually renewed re-
searches as to the site of the formation of the blood and its mode
of formation. Who among us does not feel it as a wound, a
painful raw in his scientific manhood that we still are unable to
say ' *Here* and *thus* do the red blood-corpuscles take their
origin !' " Neumann and Bĭzzozero deserve the amplest recog-
nition for their discovery that in the red marrow of bones, cells
occur with reddish-yellow homogeneous protoplasm and well-
marked nucleus, cells which accordingly are identical with the
red blood-corpuscles of the earliest period of life. These obser-
vations are easy to repeat and are fully accepted by all histolo·
gists. From these observations we know clearly *where* besides
in the spleen, we have to look for the great factory of the red
blood-corpuscles.

Hæmatogenesis is either a temporary or permanent function
of certain regions of the connective-substance apparatus of the
body, which for this purpose and during this period enters into
an open communication with the lumen of blood-vessels either
by the loss of their proper walls on the part of the capillaries
and veins, as happens in the bone-marrow or by the thinning of
their walls to such a degree, as in the spleen, that the unrestricted
in-and-out wandering of cells becomes possible. The hæmato-
genous connective-tissue becomes a sort of accessory cavity for
the lumen of the blood-vascular system. In this cavity hæmo-
globin-containing cells are produced by the conversion of
colourless cells. Professor Rindfleisch compares the process of
formation of hæmoglobin in these cells to that of fat in fat-
forming connective-tissue.

Professor Rindfleisch's special contribution to this subject
consists in : 1st, a description of the vascular plexus of the
marrow of mammalian bones which he has succeeded in injecting
(using the rib of a young guinea-pig) and of the wall-less
character of its smaller vessels. 2nd, and of greatest import-

ance, an answer to the question " How do the nucleated red corpuscles of the red bone-marrow give rise to the non-nucleated red corpuscles of the blood ?"

It is well known that this question has always been answered by hypothesis based on very slender foundation.

The old view, as to the origin of the red blood-corpuscles, was that the nucleus of certain colourless corpuscles became red and escaped as a free nucleus, the homogeneous red blood-corpuscle.

Later knowledge as to the red coloration of the whole of the mother-cell of the red corpuscle led to the assumption that the nucleus became atrophied and the whole cell converted into the non-nucleated red corpuscle. The attempts which have been made from time to time during the past few years to detect a nucleus in some form or other in the red mammalian corpuscle, point to a foregone conclusion in favour of this total conversion.

Professor Rindfleisch has, however, seen, both in embryos and more advanced individuals, the steps in the transformation of the red-coloured cell of the marrow into the non-nucleated red corpuscle which demonstrate that *the nucleus of the red coloured cell escapes and atrophies whilst the body of the cell contracts and becomes the red corpuscle.*

He gives figures of the red cells with their nuclei in the act of escaping, lying just on the limit of the cell-body or protruding from or even hanging by a mere thread to the latter. Then beside these he has seen and figures the freed nucleus and the irregular collapsed coloured body of the cell, which will soon be shaped by pressure and rolling into the disc-form of the circulating red corpuscle.

Professor Rindfleisch has endeavoured, but unsuccessfully, to witness under his own eyes the actual extrusion of a nucleus from a red cell. At the same time the intermediate series of forms observed by him are very strong evidence in favour of the view which he takes.

It seems also that Professor Rindfleisch's view is supported by certain facts of comparative anatomy which he has not himself adduced in its favour. In the Chætopodous and some other worms the *nuclei* of the vascular walls are often loosened and float in the blood as corpuscles. They are not impregnated by hæmoglobin but the plasma, in which they float, is. Whence comes the hæmoglobin of the plasma ? Clearly the cells forming the walls of the vascular system in certain regions are in the Chætopoda as in Vertebrata, hæmatogenous; in them as in Vertebrata, the body of the cell forms the hæmoglobin which in this case becomes liquid instead of retaining the form of a corpuscle,

and at the same time the nucleus is separated from the hæmo-globin-bearing body just as it is in the Mammalia, but here, as it does not there, enters into the blood stream.

Any discussion of the mode and significance of the formation of hæmoglobin in the mammalian blood, ought to take cognizance of the fact that hæmoglobin is formed in the blood of the worms above noted, in Insect larvæ, Crustacea, and even Molluscs, and further that whilst it usually occurs diffused in the plasma of the blood it does occasionally, as in the Chætopods Glycera and Capitella, the Molluscs, Solen legumen and Arca, sp., &c., take the form of special nucleated corpuscles differing from and accompanied by the usual amœboid colourless corpuscles : also it is to be noted that just as fat occurs in other cells than specialised fat-cells so do we find the muscular tissue of many Vertebrates and of some Molluscs (buccal mass) impregnated with hæmoglobin. And even in one Annelid (the sea-mouse Aphrodite) we have the cells of the nervous tissue so rich in it, that the nerve-cord is of a deep crimson colour (see ' Proc. Roy. Soc.,' No. 140, 1873).—E. RAY LANKESTER.

I. **On the Mode in which Hydra swallows its Prey.** By M. M. HARTOG, M.A., B.Sc., F.L.S., of the Owens College, Manchester. The current idea is that Hydra swallows by taking its prey in its tentacles and turning tentacles and all into its stomach. However, the part played by the tentacles ceases as soon as the mouth comes in contact with the food. The hydra then slowly stretches itself over the food in a way that recalls to some extent the manner in which a serpent " gets outside " its prey, or in which an automatic stocking might stretch itself on to the foot and leg. No care seems to be taken, however, to present the easiest point for deglutition, and an Entomostracan may be swallowed side-ways, for instance. So far are the tentacles from co-operating in the act, that they are usually reflexed away from the food ; occasionally, however, they are swung forward for a moment around the mass as if to ascertain how much remains to be swallowed.

If the prey be at all bulky, immediately after the whole act is completed the body cavity is everywhere filled and on the stretch , but after a short lapse of time the body contracts forcibly along the long axis, so that the part containing the food is globular, supported on a slender foot and with a slender apical process bearing the tentacles around the hypostome.

II. **Additional Note on Hydra.** By the Same. Since my last note I think I have found the clue to the false idea referred to. A Hydra that had swallowed a morsel larger than itself disgorged, as frequently observed, on my attempting to take it up for examination. On finding it half an hour after, three of its tentacles were

turned into its digestive cavity, whence they were successively and slowly withdrawn. As the mouth closes but slowly after disgorging, I imagine the swallowing them to have been accidental; and a similar phenomenon carelessly observed may well have given rise to a false interpretation.

It seems that here we have the true explanation of the occasional presence of nematocysts in the endoderm, and this explanation my friend, Mr. T. J. Parker, is now inclined to accept. As regards the absence of the interstitial cells from the tentacles of Hydra fusca I am not able to confirm him ; on the contrary, they are present, though in isolated patches, and not forming a continuous network as over the body. I find the best way to demonstrate these is, having killed a Hydra extended on a slide by letting fall a drop of one per cent. osmic acid on it, to at once wash away the acid by a flood of absolute alcohol, and then after a few minutes to stain with ammoniacal carmine or picro-carmine. If the Hydra is now examined in glycerine under a power sufficiently high to focus successive layers, the presence of interstitial cells can be made out. Owing to their dispersion, the want of them in a section becomes very slight evidence for their absence.

III. **On the Anal Respiration of the Copepoda.** By the Same. In a note on *Cyclops* read at the British Association I pointed out that its respiration was exclusively anal. I have now made out the same in *Canthocamptus* (fam. *Harpacticidæ*), and *Diaptomus* (fam. *Calanidæ*). In all three the mechanism is the same ; at regular intervals, after the backward sway of the intestine, the anal valves open for an instant and then close, giving just time for a slight indraught of water after the opening, a slight expulsion at the close. The necessary pressure to confine the animal seems to interfere somewhat with these movements, sometimes stopping them if excessive; hence I refrain from noting with illusory exactness the intervals between each respiratory movement.

It is to be noticed that the rectum contains as a rule liquid only, the bolus of fæces remaining in it but a short time. By endosmose the liquid in the rectum will tend to be at the same condition of gaseous saturation as the body fluid around it, kept constantly agitated by the backwards and forwards sway of the stomach. During the short interval that the anus is open an approach to gaseous equilibrium with the external water takes place, even despite the very slight movement of the water (shown by the little change of place undergone by suspended indigo or carmine particles). In the absence of any other suitable respiratory apparatus, no one can hesitate as to the function of the action I have described.

In the *Nauplius* larvæ of *Cyclops* and *Diaptomus* the working is slightly different. The rectum is a subspherical muscular sac, which at regular intervals contracts so as to leave a linear cavity (along the long axis of the animal), and immediately dilates, sucking up the water from without.

An anal respiration, such as that of *Cyclops,* is found widely among *Crustacea*—even those which have well developed gills like *Astacus,* which is one of the highest forms. It has been demonstrated in *Phyllopoda* and *Cladocera,* and is probably the exclusive mode in *Leptodora,* as shown by Weismann. That it is therefore primitive, and should be expected to occur in the primitive or at least very generalised group of the *Copepoda,* is an obvious deduction. Hence I anticipate that the homœomorphic *zoœa* larvæ of the *Decapoda* will prove to have this same mode of respiration.

If there be any connection between Rotifers and *Nauplius,* it is easy to make out the origin of the arrangement in the latter. The ciliated funnels and lateral canals of the former can only be of service when there is a thin unchitinised anterior surface through which water can transude into the cœlom; by the extension of chitinisation over the whole surface these organs lose their function and abort, while the cloacal "contractile vesicle" takes on an inspiratory as as well as an expiratory function, and becomes more or less confounded with the rectum, from which probably, even in Rotifers, it takes origin.

Here must be noticed the wide diffusion of anal respiration in aquatic Insect larvæ (alternate inspiration and expiration by the pumping movements of the rectum). This would point to a common origin with Crustacea.

A list of the groups in which anal respiration is made out may be added.

  Vermes :

      Rotifera.
      Gephyrea.
      Oligochæto Limicola.

  Echinodermata.

      Holothuroidea.

  Arthropoda.

      Crustacea (general).
      Insecta (most aquatic larvæ).

  Mollusca.

      Dentalium.

Dr. G. von Koch's Method of Preparing Sections of Corals.—When working at the structure of corals during the Challenger expedition I found very great difficulty in determining the exact relations of the hard to the soft parts. It is

easy enough to prepare sections of the soft tissues after decalci-
fication, and of the hard dried corallum by the old process of
grinding, but the results thus obtained have then to be com-
bined more or less by guess work.   Dr. G. von Koch has lately
sent me a series of microscopic sections of corals prepared by
him by means of a method which he described in the 'Zoolo-
gischer Anzeiger,' No. 2, 1878, S. 36.   In these sections the
hard and soft tissues are maintained in their exact relations to
one another, and both are reduced to a sufficient thinness to
exhibit their minute structure in all essential details.   Amongst
them, for example, is a complete section across a Caryophyllia, in
which the arrangement of the mesenteries with regard to the
septa is most fully and clearly exhibited.   Dr. von Koch has
described the results at which he has arrived by means of his
method of section cutting, in a series of papers published in the
'Morphologisches Jahrbuch,' and elsewhere.   His latest paper,
which gives an account of some points in the structures of
Caryophyllia, is contained in the 'Morphol. Jahrb.,' Bd. v, S.
316 ("Bemerkungen über das Skelett der Korallen").

My object in writing this note is to testify to the great success
of Dr. von Koch's method.   It will yield valuable results not
only in the case of corals, but also in all other problems of
histology or minute anatomy in which the relations of hard and
soft parts have to be determined.   It might, perhaps, be em-
ployed with advantage in the examination of the structure of
Corti's organ.   Sections could thus be prepared of the undecal-
cified cochlea in which the components of the organ of Corti
would be seen *in situ*, and unaltered by the action of acids.
Sections of injected bone, showing the relations of the blood-
vessels to the Haversian system, could also thus be made.
Sections across the arms of undecalcified Crinoids and Starfish,
and many similar preparations, suggest themselves as likely to
yield valuable results.   Dr. von Koch's method is described in
full in the 'Zoologischer Anzeiger' in the notice quoted above.
The hardened objects of which sections are to be cut are stained
and treated with absolute alcohol, and then placed in a solution
of gum copal in chloroform.   The objects are then slowly dried
by means of artificial heat till they are stony hard.   They are
then cut into sections with a fine saw, and the sections are
rubbed smooth on a bone on one side, then fastened to the slide
with the copal solution, and ground down with a grindstone
and hone on the other, just as in the case of ordinary sections of
bones and teeth.—H. N. MOSELEY.

MEMOIRS.

On the STRUCTURE and HOMOLOGIES of the GERMINAL
LAYERS of the EMBRYO.[1]   By F. M. BALFOUR, M.A.,
F.R.S., Fellow of Trinity College, Cambridge.

THE discovery by Pander and Von Baer that the young
embryos of vertebrated animals were formed of distinct
layers of cells known as the germinal layers, and that there
were special and constant relations between these layers and
the adult organs throughout the group, remained for a long
time an isolated fact.   In 1859 Huxley made an important
step towards the explanation of the nature of these layers
by comparing them with the ectoderm and endoderm of the
Hydrozoa.

In spite of Huxley's comparison it was for a long time
the generally accepted view that germinal layers similar
to those in the Vertebrata were not found amongst inverte-
brated animals.   The brilliant researches of Kowalewsky on
the development of a great variety of invertebrate forms
first proved that the accepted view was erroneous, and they
led Lankester (No. 16) and Haeckel (No. 9) to publish
certain speculations which have had an incalculable
influence in stimulating and directing modern embryological
research.[2]

At the time when the essays containing these speculations
were published there appeared to be every probability of
a definite answer being given to the questions raised in
them.   The results of extended investigations during the
last few years have, however, shown that these expectations
were premature; and there are very few embryologists who

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[1] This paper, with some modifications, forms part of a chapter in the
forthcoming volume of my treatise on ' Comparative Embryology.'

[2] If I do not refer to the above authors to any very great extent in the
sequel, it is not because I undervalue the importance of their work, but
because their views have been so much before the public that it is quite
unnecessary to discuss them in detail.

would venture to assert that any hypotheses, which can now
be put forward as to the mode of origin or the homologies of
the germinal layers, have more than a tentative value.

In the following pages I aim more at summarising the
facts, and critically examining the different theories which
can be held, than at dogmatically supporting any definite
views of my own.

In all the Metazoa the development of which has been in-
vestigated the first process of differentiation which follows
upon the segmentation consists in the cells of the organism be-
coming divided into two groups or layers, known respectively
as epiblast and hypoblast.[1]   This process may commence
during later phases of the segmentation, but is generally not
completed till after the close of the segmentation.   Not
only do the cells of the blastoderm become differentiated
into two layers, but these two layers, in the case of a very
large number of ova with but little food-yolk, constitute a
double-walled sack—the gastrula (fig 1)—the characters of
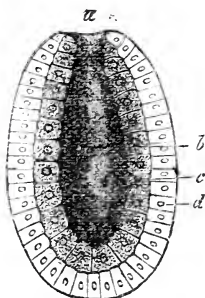
FIG. 1.—*Diagram of a Gastrula.*   (From Gegenbaur.)   *a.* Mouth; *b.*
    archenteron; *c.* hypoblast; *d.* epiblast.

which are too well known to require further description.
It is generally admitted, and will be assumed in the sequel,
that the segmented ovum represents phylogenetically a
compound Protozoon; and following the same lines of
phylogenetic speculation, it may be concluded that the two-
layered condition of the organism represents in a general
way the passage from the Protozoon to the Metazoon
condition.   It is probable that we may safely go further,
and assert that the gastrula reproduces, with more or less
fidelity, a stage in the evolution of the Metazoa, permanent
in the simpler Hydrozoa, during which the organism was

[1] In a few cases amongst the Mollusca and Chætopoda, &c., the meso-
blast is differentiated simultaneously with the two other layers.   These
cases may for the moment be left out of consideration.

provided with (1) a fully developed digestive cavity (fig. 1 b) lined by the secretory hypoblast, (2) an oral opening (a), and (3) a superficial epiblast (d). These generalisations, which are now widely accepted, are no doubt very valuable, but they leave unanswered the following important questions :

(1) By what steps did the compound Protozoon become differentiated into a Metazoon ?

(2) Are there any grounds for thinking that there is more than one line along which the Metazoa have become independently differentiated from the Protozoa ?

(3.) To what extent is there a complete homology between the two primary germinal layers throughout the Metazoa ?

Ontologenetically there are a great variety of processes by which the passage from the segmented ovum to the two-layered or diploblastic condition is arrived at.

These processes may be grouped under the following heads :

1. **Invagination.**—Under this term a considerable number of closely connected processes are included. When the segmentation results in the formation of a blastosphere, one half of the blastosphere may be pushed in towards the opposite half, and a gastrula be thus produced (fig. 2, A and B). This process is known as embolic invagination. Another
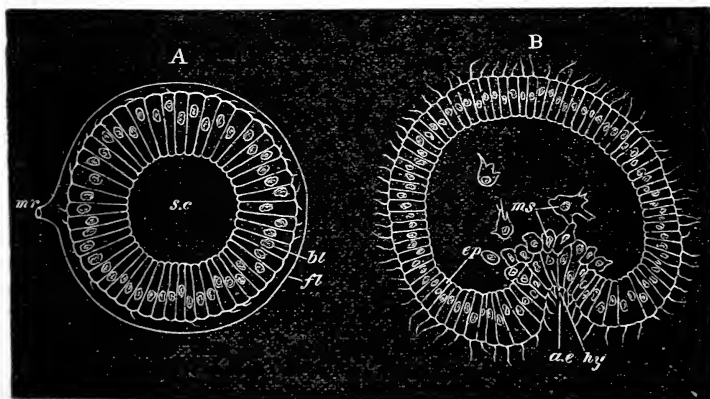
FIG. 2.—*Two Stages in the Development of Holothuria tubulosa, viewed in optical section.* (After Selenka.) A. Stage at the close of the segmentation. B. Gastrula stage. *mr.* micropyle ; *fl.* chorion ; *s. c.* segmentation cavity ; *bl.* blastoderm ; *ep.* epiblast ; *hy.* hypoblast ; *ms.* amœboid cells derived from hypoblast ; *a.e.* archenteron.

process, known as epibolic invagination, consists in epiblast cells growing round and enclosing the hypoblast (fig. 3). This

process replaces the former process when the hypoblast cells are so bulky from being distended by food-yolk that their invagination is mechanically impossible.
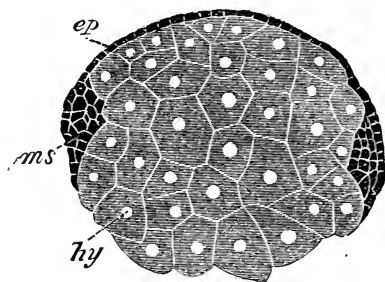
FIG. 3.—*Transverse Section through the Ovum of Euaxes during an early stage of development to show the nature of epibolic invagination.* (After Kowalevsky.) *ep.* epiblast; *ms.* mesoblastic band; *hy.* hypoblast.

There are various peculiar modifications of invagination which cannot be dealt with in detail.

Invagination in one form or other occurs in some or all the members of the following groups :

The Dicyemidæ, Calcispongiæ (after the amphiblastula stage) and Silicispongiæ, Cœlenterata, Turbellaria, Nemertea, Rotifera, Mollusca, Polyzoa, Brachiopoda, Chætopoda, Discophora, Gephyrea, Chætognatha, Nematelminthes, Crustacea, Echinodermata, and Chordata.

The gastrula of the Crustacea is peculiar, as is also that of many of the Chordata (Reptilia, Aves, Mammalia), but there is every reason to suppose that the gastrulæ of these groups are simply modifications of the normal type.

2. **Delamination.**—Three types of delamination may be distinguished :

a. Delamination where the cells of a solid morula become divided into a superficial epiblast, and a central solid mass in which the digestive cavity is subsequently hollowed out (fig. 4).

b. Delamination where the segmented ovum has the form of a blastosphere, the cells of which give rise by budding to scattered cells in the interior of the vesicle, which, though they may at first form a solid mass, finally arrange themselves in the form of a definite layer around a central digestive cavity (fig. 5), or, in the case of some sponges, around several cavities.

c. Delamination where the segmented ovum has the form
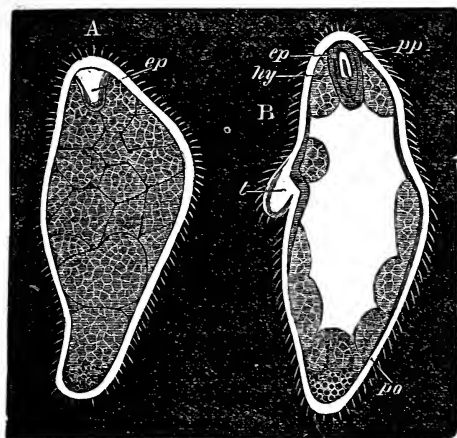
FIG. 4.—*Two Stages in the Development of Stephanomia pictum, to illustrate delamination.* (After Metschnikoff.) A. Stage after the delamination; *ep.* epiblastic invagination to form pneumatocyst. B. Later stage after the formation of the gastric cavity in the solid hypoblast. *po.* polypite; *t.* tentacle; *pp.* pneumatocyst; *ep.* epiblast of pneumatocyst; *hy.* hypoblast surrounding pneumatocyst.

of a blastosphere in the cells of which the protoplasm is differentiated into an inner and an outer part. By a subsequent
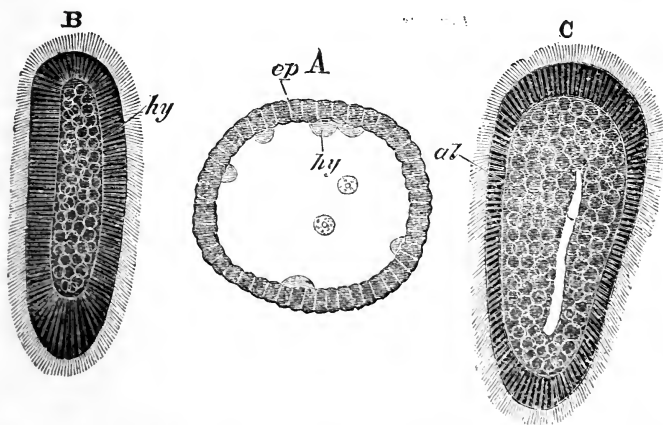
FIG. 5.—*Three Larval Stages of Eucope polystyla.* (After Kowalevsky.) A. Blastosphere stage with hypoblast spheres becoming budded off into central cavity. B. Planula stage with solid hypoblast. C. Planula stage with a gastric cavity. *ep.* epiblast; *hy.* hypoblast; *al.* gastric cavity.

process the inner parts of the cells become separated from

the outer, and the walls of the blastosphere are so divided
into two distinct layers (fig. 8).

Although the third of these processes is usually regarded as
the type of delamination, it does not, so far as I know,
occur in nature, but is most nearly approached in Geryonia
(fig. 6).

The first type of delamination is found in the Cerato-
spongiæ, some Silicispongiæ (?), and in many Hydrozoa and
Actinozoa, and in Nemertea and Nematelminthes (*Gordi-
oidea ?*). The second type occurs in many Porifera [*Calci-
spongiæ (Ascetta), Myxospongiæ*], and in some Cœlenterata,
and Brachiopoda (*Thecidium*).
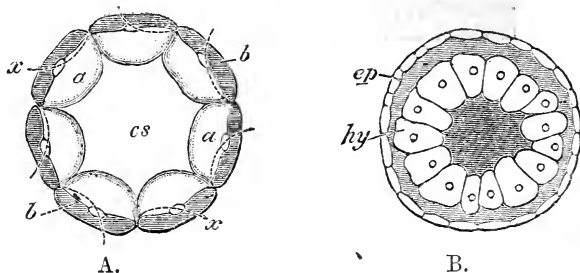
Fig. 6.—*Diagramatic Figures showing the Delamination of the Embryo
of Geryonia.* (After Fol.) A. Stage at the commencement of the
delamination; the dotted lines show the course of the next planes of
division. B. Stage at the close of the delamination. *cs.* segmentation
cavity; *a.* endoplasm; *b.* ectoplasm.

Delamination and invagination are undoubtedly the two
most frequent modes in which the layers are differentiated, but
there are in addition several others. In the first place the
whole of the Tracheata (with the apparent exception of the
Scorpion) develop, so far as is known, on a plan of their
own, which approaches delamination. This consists in the
appearance of a superficial layer of cells enclosing a central
yolk mass, which corresponds to the hypoblast (figs. 7 and
17). This mode of development might be classed under
delamination were it not for the fact that the early develop-
ment of the Crustacea is almost the same, but is subsequently
followed by an invagination (fig. 11), which apparently
corresponds to the normal invagination of other types.
There are grounds for thinking that the tracheate type of
formation of the epiblast and hypoblast is a secondary
modification of an invaginate type.

The type of some Turbellaria (*Stylocopsis ponticus*) and

that of Nephelis amongst the Discophora is not capable of being reduced to the invaginate type.
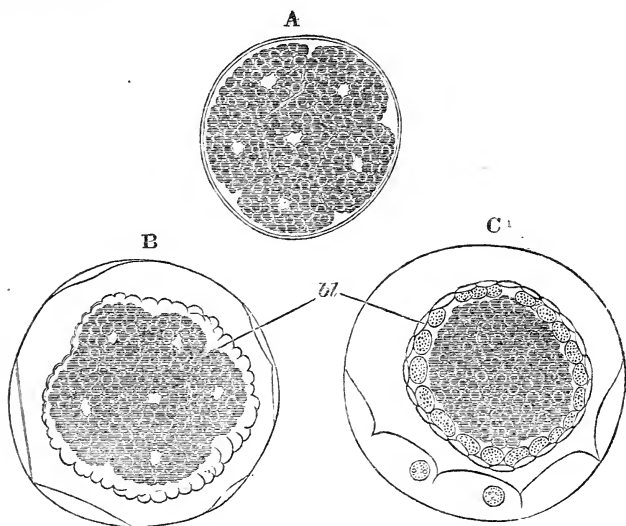
FIG. 7.—*Segmentation and Formation of the Blastoderm in Chelifer.* (After Metschnikoff.) In A the ovum is divided into a number of separate segments. In B a number of small cells have appeared (*bl.*), which form a blastoderm enveloping the large yolk spheres. In C the blastoderm has become divided into two layers.

The development of almost all the parasitic groups, *i.e.* the Trematoda, the Cestoda, the Acanthocephala, and the Linguatulida, and also of the Tardigrada, Pycnogonida, and other minor groups, is too imperfectly known to be classed with either the delaminate or invaginate types.

It will, I think, be conceded on all sides that, if any of the ontogenetic processes by which a gastrula form is reached are repetitions of the process by which a single two-layered gastrula was actually developed from a compound Protozoon, these processes are most probably either invagination or delamination.

The much disputed questions which have been raised about the gastrula and planula theories, originally put forward by Haeckel and Lankester, resolve themselves then into the single question, whether any, and if so which, of the ontogenetic processes by which the gastrula is formed are repetitions of the phylogenetic origin of the gastrula.

It is very difficult to bring forward arguments of a conclusive kind in favour of either of these processes. The fact that delaminate and invaginate gastrulæ are in several

instances found coexisting in the same group renders it certain that there are not two independent phyla of the Metazoa, derived respectively from an invaginate and a delaminate gastrula.[1]

The four most important cases in which the two processes coexist are the Porifera, the Cœlenterata, the Nemertea, and the Brachiopoda. In the cases of the Porifera and Cœlenterata, there do not appear to me to be any means of deciding which of these processes is derived from the other; but in the Nemertea and the Brachiopoda the case is different. In all the types of Nemertea in which the development is relatively not abbreviated there is an invaginate gastrula, while in the types with a greatly abbreviated development there is a delaminate gastrula. It would seem to follow from this that a delaminate gastrula has here been a secondary result of an abbreviation in the development. In the Brachiopoda, again, the majority of types develop by a process of invagination, while Thecidium appears to develop by delamination; here, also, the delaminate type would appear to be secondarily derived from the invaginate.

If these considerations are justified, delamination must be in some instances secondarily derived from invagination; and this fact is so far an argument in favour of the more primitive nature of invagination; though it by no means follows that invagination contains a repetition of the steps by which the Metazoa were derived from the Protozoa.

It does not, therefore, seem possible to decide conclusively in favour of either of these processes by a comparison of the cases where they occur in the same groups.

The relative frequency of the two processes supplies us with another possible means for deciding between them; and there is no doubt that here again the scale inclines towards invagination. It must, however, be borne in mind that the frequency of the process of invagination admits of another possible explanation. There is a continual tendency for

[1] It is not difficult to picture a possible derivation of delamination from invagination, while a comparison of the formation of the inner layers (mesoblast and hypoblast) in Ascetta (amongst the sponges) and in the Echinodermata shows a very simple way in which it is possible to conceive of a passage of delamination into invagination. In Ascetta the cells, which give rise to the mesoblast and hypoblast, are budded off from the inner wall of the blastosphere, especially at one point; while in Echinodermata (fig. 2) there is a small invaginated sac which gives rise to the hypoblast, while from the walls of this sac amœboid cells are budded off which give rise to the mesoblast. If we suppose the hypoblast cells budded off at one point in Ascetta gradually to form an invaginated sac, while the mesoblast cells continued to be budded off as before, we should pass from the delaminate type of Ascetta to the invaginate type of an Echinoderm.

the processes of development to be abbreviated and simpli-
fied, and it is quite possible that the frequent occurrence
of invagination is due to the fact of its being, in most
cases, the simplest means by which the two-layered condition
can be reached. But this argument can have but little weight
until it can be shown that invagination is a simpler process
than delamination.

If it were the case that the blastopore had *in all types*
the same relation to the adult mouth, there would be strong
grounds for regarding the invaginate gastrula as an ancestral
form; but the fact that this is by no means so is an argu-
ment of great weight in favour of some other explanation
of the frequency of invagination.

The force of this consideration can best be displayed by a
short summary of the fate of the blastopore in different forms.

The fate of the blastopore is so variable that it is diffi-
cult even to classify the cases which have been described.

The forms which have been classed together under the

(1) It becomes the permanent mouth in the following forms :[1]
  Cœlenterata.—Pelagia, Cereanthus.
  Turbellaria.—Leptoplana (?), Thysanozoon.
  Nemertea.—Pilidium, larvæ of the type of Desor.
  Mollusca.—In numerous examples of most molluscan groups, ex-
    cept the Cephalopoda.
  Chætopoda.—Most Oligochæta, and probably many Polychæta.
  Gephyrea.—Phascolosoma, Phoronis.
  Nematelminthes.—Cucullanus.
(2) It closes in the position where the mouth is subsequently formed.
  Cœlenterata.—Ctenophora (?).
  Mollusca.—In numerous examples of most Molluscan groups, except
    the Cephalopoda.
  Crustacea.—Cirripedia (?), some Cladocera (Moina) (?).
(3) It becomes the permanent anus.
  Mollusca.—Paludina.
  Chætopoda.—Serpula and some other types.
  Echinodermata.—Almost universally, except amongst the Crinoidea.
(4) It closes in the position where the anus is subsequently formed.
  Echinodermata.—Crinoidea.
(5) It closes in a position which does not correspond or is not known
to correspond[2] either with the future mouth or anus.—*Porifera :* Sycandra.
Cœlenterata : Chrysaora,* Aurelia.* *Nemertea :** some larvæ which develop
without a metamorphosis. *Rotifera.** *Mollusca :* Cephalopoda. *Polyzoa.**
Brachiopoda : Argiope, Terebratula, Terebratulina. *Chætopoda :* Euaxes.
Discophora : Clepsine. *Gephyrea :* Bonellia.* *Chætognatha. Crustacea :*
Decapoda. *Chordata.*

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[1] The above list is somewhat tentative ; and future investigations will
probably show that many of the statements at present current about the
position of the blastopore are inaccurate.

[2] The forms in which the position of the blastopore in relation to the
mouth or anus is not known are marked with an asterisk.

last heading vary considerably in the character of the blastopore. In some cases the fact of its not coinciding either with the mouth or anus appears to be due simply to the presence of a large amount of food-yolk. The cases of the Cephalopoda, of Euaxes, and perhaps of Clepsine and Bonellia, are to be explained in this way: in the case of all these forms, except Bonellia, the blastopore has the form of an elongated slit along the ventral surface. This type of blastopore is characteristic of the Mollusca generally, of the Polyzoa, of the Nematelminthes, and very possibly of the Chætopoda and Discophora. In the Brachiopoda and the Chætognatha (fig. 12 B) the blastopore is situated, so far as can be determined, behind the future anus. In many Decapoda, amongst the Crustacea, the blastopore is placed behind, but not far from, the anus. In the Chordata it is also placed posteriorly, and, remarkably enough, remains, in a large number of forms, for some time in connection with the neural tube by a neurenteric canal.

The great variations in the character of the gastrula, indicated in the above summary, go far to show that if the gastrulæ, as we find them in most types, have any ancestral characters, these characters can only be very general ones. This may best be shown by the consideration of a few striking instances. The blastopore in Mollusca has an elongated slit-like form, extending along the ventral surface from the mouth to the anus; in Echinodermata it is a narrow pore, remaining as the anus. In most Chætopoda it is a pore remaining as the mouth, but in some as the anus. In Chordata it is a posteriorly-placed pore, opening into the neural canal.

It is clearly out of the question to explain these differences in connection with the characters of ancestral forms. They can only be accounted for as secondary adaptations for the convenience of development.

The epibolic gastrula of Mammalia is a still more striking case of a secondary embryonic process, and is not directly derived from the gastrula of the lower Chordata. It probably originated in connection with the loss of food-yolk which took place on the establishment of a placental nutrition for the fœtus. The epibolic gastrula of the Scorpion, of Isopods, and of other Arthropoda, seems also to be a derived gastrula. These instances of secondary gastrulæ are very probably by no means isolated, and should serve as a warning against laying too much stress upon the frequency of the occurrence of invagination. The great influence of the food-

yolk upon the early development might be illustrated by numerous examples, especially amongst the Chordata.[1]

If the descendants of a form with a large amount of food-yolk in its ova were to have ova with but little food-yolk, the type of formation of the germinal layers which would thereby result would be by no means the same as that of the ancestors of the forms with much food-yolk, but would probably be something very different, as in the case of Mammalia. Yet amongst the countless generations of ancestors of most existing forms, such oscillations in the amount of the food-yolk must have occurred in a large number of instances.

The whole of the above considerations point towards the view that the formation of the hypoblast by invagination, as it occurs in most forms at the present day, can have no special phylogenetic significance, and that the argument of frequency, in favour of invagination as opposed to delamination, is not of prime importance.

A third possible method of deciding between delamination and invagination is to be found in the consideration as to which of these processes occurs in the most primitive forms. If there were any agreement amongst primitive forms as to the type of their development this argument might have some weight. On the whole, delamination is, no doubt, characteristic of the most primitive types, but the not infrequent occurrence of invagination in both the Cœlenterata and the Porifera—the two groups which would on all hands be admitted to be amongst the most primitive—deprives this argument of much of the value it might otherwise have.

To sum up : in the present state of our knowledge there are no satisfactory data for deciding which of the two processes is the more primitive ; nor, considering the almost indisputable fact that both these processes have in many instances had a purely secondary origin, can any valid arguments be produced to show that either of them reproduces the mode of passage between the Protozoa and the ancestral two-layered Metazoa. These conclusions do not, however, throw any doubt upon the fact that the gastrula, however evolved, was a primitive form of the Metazoa ; since this conclusion is founded upon the actual existence of adult gastrula forms independently of their occurrence in development.

Though embryology does not at present furnish us with an answer to the question how the Metazoa became developed

[1] *Vide* F. M. Balfour, " A comparison of the early stages in the development of Vertebrates," ' Quart. Journ. Micr. Sci.,' vol. 1875.

from the Protozoa, it is nevertheless worth while reviewing some of the processes by which this can be conceived to have occurred.

On purely *à priori* grounds there is in my opinion more to be said for invagination than for any other view.

On this view we may suppose that the colony of Protozoa in the course of conversion into Metazoa had the form of a blastosphere ; and that at one pole of this a depression appeared. The cells lining this depression we may suppose to have been amœboid, and to have carried on the work of digestion ; while the remaining cells were probably ciliated. The digestion may be supposed to have been at first carried on in the interior of the cells, as in the Protozoa ; but, as the depression became deeper (in order to increase the area of nutritive cells and to retain the food) a digestive secretion probably became poured out from the cells lining it, and the mode of digestion generally characteristic of the Metazoa was thereby inaugurated. It may be noted that an intra-cellular protozoon type of digestion persists in the Porifera, and appears also to occur in many Cœlenterata, Turbellaria, &c., though in most of these cases both kinds of digestion probably occur simultaneously.[1]

Another hypothetical mode of passage which fits in with delamination has been put forward by Lankester, and is illustrated by fig. 8. He supposes that at the blastosphere stage the fluid in the centre of the colony acquired special digestive properties; the inner ends of the cells had at this stage somewhat different properties to the outer, and the food was still incepted by the surface of the cells (fig. 8, $_3$). In a later stage of the process the inner portions of the cells became separated off as the hypoblast, while the food, though still ingested in the form of solid particles by the superficial cells, was carried through the protoplasm into the central digestive cavity. Later (fig. 8, $_4$), the point where the food entered became localised, and eventually a mouth became formed at this point.

The main objection which can be raised against Lankester's view is that it presupposes a type of delamination which does not occur in nature except in Geryonia.

Metschnikoff has propounded a third view with reference to delamination. He starts as before with a ciliated blastosphere. He next supposes the cells from the walls of this to become budded off into the central cavity, as in Eucope

[1] J. Parker, "On the Histology of *Hydra fusca*," 'Quart. Journ. Micr. Science ;' and El. Metschnikoff, " Ueb. die Intracelluläre Verdauung bei Cœlenteraten," ' Zoologischer Anzeiger,' No. 56, vol. iii, 1880.

(fig. 5), and to lose their cilia. These cells give rise to an internal parenchyma, which carries on an intra-cellular di-
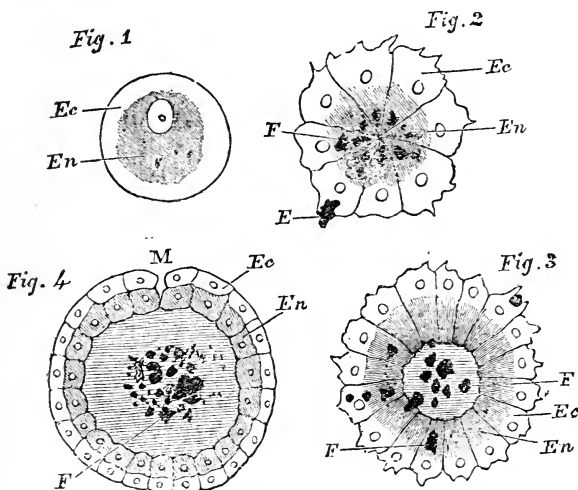
FIG. 8.—*Diagram showing the Formation of a Gastrula by Delamination.* (From Lankester.) Fig. 1, ovum; fig. 2, stage in segmentation; fig. 3, commencement of delamination after the appearance of a central cavity; fig. 4, delamination completed, mouth forming at *M*. In figs. 1, 2, and 3, *Ec.* is ectoplasm, and *En.* is endoplasm. In fig. 4, *Ec.* is epiblast, and *En.* hypoblast. *F.* food particles.

gestion. At a later stage a central digestive cavity is supposed to be formed. This view of the passage from the protozoon to the metazoon state, though to my mind improbable in itself, fits in very well with the ontogeny of the lower Hydrozoa.

Another view has been put forward by myself,[1] to the effect that the amphiblastula larva of Calcispongiæ may be a transitional form between the Protozoa and the Metazoa, composed of a hemisphere of nutritive amœboid cells and a hemisphere of ciliated cells. The absence of such a larval form in the Cœlenterata and higher Metazoa is opposed, however, to this larva being regarded as a transitional form, except for the Porifera.

It is obvious that so long as there is complete uncertainty as to the value to be attached to the early developmental processes, it is not possible to decide from these processes whether there is only a single Metazoon phylum or whether there may not be two or more such phyla. At

[1] F. M. Balfour, 'A Treatise on Comparative Embryology,' vol. i, p. 122.

the same time there appear to be strong arguments for re-
garding the Porifera as a phylum of the Metazoa derived
independently from the Protozoa. This seems to me to be
shown (1) by the striking larval peculiarities of the Porifera;
(2) by the early development of the mesoblast in the Porifera,
which stands in strong contrast to the absence of this layer in
the embryos of most Cœlenterata; and above all, (3) by the re-
markable characters of the system of digestive channels. A
further argument in the same direction is supplied by the
fact that the germinal layers of the Sponges very probably
do not correspond physiologically to the germinal layers of
other types. Whether or no the amphiblastula larva is, as
suggested above, to be regarded as the larval ancestor of the
Porifera must be left as an open question.

The question as to how far there is a complete homology
between the two primary germinal layers throughout the
Metazoa was the third of the questions propounded.

Since there are some Metazoa with only two germinal
layers, and other Metazoa with three, and since, as is shown in
the following section, the third layer or mesoblast can only be
regarded as a derivative of one or both the primary layers,
it is clear that a complete homology between the two
primary germinal layers does not exist.

That there is a general homology appears on the other
hand hardly open to doubt.

The primary layers are usually continuous with each other,
near one or both (when both are present) the openings of
the alimentary tract.

As a rule an oral and anal section of the alimentary tract
—the stomodæum and proctodæum—are derived from the
epiblast; but the limits of both these sections are so
variable, sometimes even in closely allied forms, that it
is difficult to avoid the conclusion that there is a border-
land between the epiblast and hypoblast, which appears
by its development to belong in some forms to the epi-
blast and in other forms to the hypoblast. If this is not
the case it is necessary to admit that there are instances in
which a very large portion of the alimentary canal is formed
of epiblast. In some of the Isopods, for example, the
stomodæum and proctodæum give rise to almost the whole
of the alimentary canal with its appendages, except the
liver.

The origin of the Mesoblast.—The diploblastic condition of
the organism preceded, as we have seen, the triploblastic.
The epiblast during the diploblastic condition was, as appears
from such forms as Hydra, especially the sensory and pro-

tective layer, while the hypoblast was the secretory layer; both layers giving rise to muscular elements. It must not, however, be supposed that in the early diploblastic ancestors there was a complete differentiation of function, but there is reason to think that both the primary layers retained an indefinite capacity for developing into any form of tissue.[1] The fact of the triploblastic condition being later than the diploblastic proves in a conclusive way that the mesoblast is a derivate of one or both the primary layers. In the Cœlenterata we can study the actual origin from the two primary layers of various forms of tissue which in the higher types become the mesoblast.[2] This fact, as well as general *à priori* considerations, conclusively prove that *the mesoblast did not at first originate as a mass of independent cells between the two primary layers, but that in the first instance it arose as histological differentiations of the two layers, and that its condition in the embryo as an independent layer of undifferentiated cells is a secondary condition, brought about by the general tendency towards a simplification of development, and a retardation of histological differentiation.*[3]

In addition to the probably degraded Dicyemidæ and Orthonectidæ, the Cœlenterata are the only group in which a completely differentiated mesoblast is not always present. In other words, the Cœlenterata are the only group in which there is not found in the embryo an undifferentiated mass of cells from which the majority of the organs situated between the epidermis and the alimentary epithelium are dveloped.

[1] The Hertwigs (No. 13) have shown that nervous structures are developed in the hypoblast in the Actinozoa.

[2] There is considerable confusion in the use of the names for the embryonic layers. In some cases various tissues formed by differentiations of the primary layers have been called mesoblast. Schultze, and more recently the Hertwigs, have pointed out the inconvenience of this nomenclature. In some of the Cœlenterata it is difficult to decide in certain instances (*e. g.* Symposium) whether the cells which give rise to a particular tissue of the adult are to be regarded as forming a mesoblast, *i. e.* a middle undifferentiated layer of cells, or arise as already histologically differentiated elements from one of the primary layers. The attempt to distinguish by a special nomenclature the epiblast and hypoblast after and before the separation of the mesoblast, which has been made by Allen Thomson (No. 1), appears to me inconvenient in practice. A proposal of the Hertwigs to adopt special names for the outer and inner limiting membranes of the adult, and for the interposed mass of organs, appears to me unnecessary, and only likely to introduce confusion into an already complicated nomenclature.

[3] The causes which give rise to a retardation of histological differentiation will be dealt with in a sequel to this paper treating of larval characters and larval forms.

The organs invariably derived, in the triploblastic forms, from the mesoblast, are the vascular and lymphatic systems, the muscular system, and the greater part of the connective tissue and the excretory system. On the other hand, the nervous system and organs of sense (with a few possible exceptions), the epithelium of most glands, and a few exceptional connective-tissue organs, *i. e.* the notochord, are developed from the two primary layers.

The fact of the first-named set of organs being invariably derived from the mesoblast, points to the establishment of the two following propositions :— (1) *That with the differentiation of the mesoblast as a distinct layer by the process already explained, the two primary layers lost for the most part the capacity they primitively possessed of giving rise to muscular and connective-tissue differentiations.* (2) *That the mesoblast throughout the triploblastic Metazoa, in so far as these forms have sprung from a common triploblastic ancestor, is an homologous structure.*

The second proposition follows from the first. The mesoblast can only have ceased to be homologous throughout the triploblastica by additions from the two primary layers, and the existence of such additions is negatived by the first proposition.

These two propositions, which hang together, are possibly only approximately true. In the first place, it is quite possible that fresh differentiations from the two primary layers may have arisen after the triploblastic condition had been established, and by the process of simplification of development and precocious segregation, as Lankester calls it, have become merged in the normal mesoblast ; or such differentiations may have taken place in forms, the development of which has not yet been investigated. Had this, however, been a frequent occurrence, it is hardly likely that no instance of it should have been recorded for the muscular system and connective tissue ;[1] so that it is probable that the muscular system of all existing triploblastic forms has been differentiated from the muscular system of the ancestor of the triploblastica. In the case of other tissues there are a few instances which might be regarded as examples of an organ primitively developed in one of the two primary layers having become secondarily carried into the mesoblast. The notochord has sometimes been cited as such an organ, but it now appears probable that its hypoblastic origin can always be demonstrated. The de-

[1] The connective-tissue test of the Tunicata, though derived from the epiblast, is not really an example of such a differentiation.

velopment of the generative organs in the Invertebrata is still very imperfectly known, but it is possible, that although usually developed in the mesoblast, they sometimes (*e. g.* Insecta) retain their primitive development from the epiblast.

The nervous system, although imbedded in mesoblastic
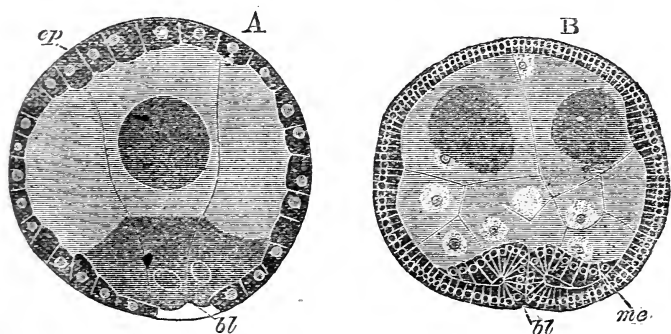
FIG. 9.—*Epibolic Gastrula of Bonellia.* (After Spengel.) A. Stage when the four hypoblast cells are nearly enclosed. B. Stage after the formation of the mesoblast has commenced by an infolding of the lips of the blastopore. *ep.* Epiblast; *me.* mesoblast; *bl.* blastopore.

derivates in the adults of all the higher triploblastica, retains with marvellous constancy its epiblastic origin (though it is usually separated from the epiblast prior to its histogenic differentiation); yet in the Cephalopoda, and some other Mollusca, the evidence is in favour of its developing in the mesoblast. Should future investigations confirm these conclusions, a good example will be afforded of an organ changing the layer from which it develops. The explanation of such a change would be precisely the same as that already given for the mesoblast as a whole.

The actual mode of origin of various tissues, which in the true triploblastic forms constitute the mesoblast, can be traced in the Cœlenterata.[1] In this group the epiblast and hypoblast both give rise to muscular and connective-tissue elements; and although the main part of the nervous system is formed in the epiblast, it seems certain that in some types nerves may be derived from the hypoblast.[2] These facts are extremely in-

[1] The reader is referred for this subject to the extremely valuable memoirs which have been recently published by the Hertwigs, especially (No. 13). He will find a general account of the subject written before the appearance of the Hertwigs' memoir in p. 149 and 150 of volume I of my treatise on 'Comparative Embryology.'

[2] It would be interesting to know about the history of the various nervous structures found in the walls of the alimentary tract in the higher forms. I have shown ('Development of Elasmobranch Fishes,' p. 172) that the central part of the sympathetic system is derived from the epiblast.

teresting, but it is by no means certain that any conclusions can be directly drawn from them as to the actual origin of the mesoblast in the triploblastic forms, till we know from what diploblastic forms the triploblastica originated. All that they show is that any part of the mesoblast may have originated from either of the primitive layers.
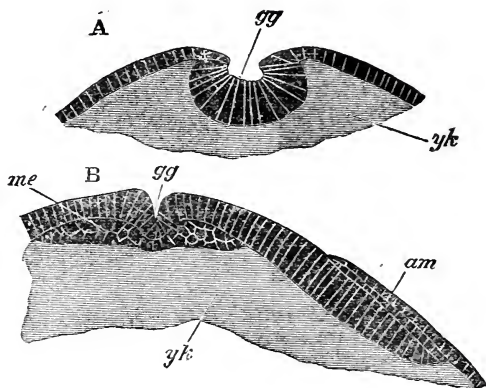
Fig. 10.—*The Transverse Sections through Embryos of Hydrophilus piceus.* (After Kowalevsky.) A. Section through an embryo at the point where the two germinal folds most approximate. B. Section through an embryo, in the anterior region where the folds of the amnion have not united. *gg.* germinal groove; *me.* mesoblast; *am.* amnion; *yk.* yolk.

For further light as to the origin of the mesoblast, it is necessary to turn to its actual development.

The following summary illustrates the more important modes in which the mesoblast originates.

1. It grows inwards from the lips of the blastopore as a pair of bands. In these cases it may originate (1) from cells which are clearly hypoblastic, (2) from cells which are clearly epiblastic, (3) from cells which cannot be regarded as belonging to either layer.

Mollusca.—Gasteropoda, Cephalopoda, and Lamellibranchiata. In Gasteropoda and Lamellibranchiata it sometimes originates from a pair of cells at the lips of the blastopore, though very probably some of the elements subsequently come from the epiblast; and in Cephalopoda as a ring of cells round the edge of the blastoderm.

Polyzoa Entoprocta.—It originates from a pair of cells at the lips of the blastopore.

Chætopoda.—Euaxes. It arises as a ridge of cells at the lips of the blastopore (fig. 3).

Gephyrea.—Bonellia. It arises (fig. 9) as an infolding of the epiblastic lips of the blastopore.
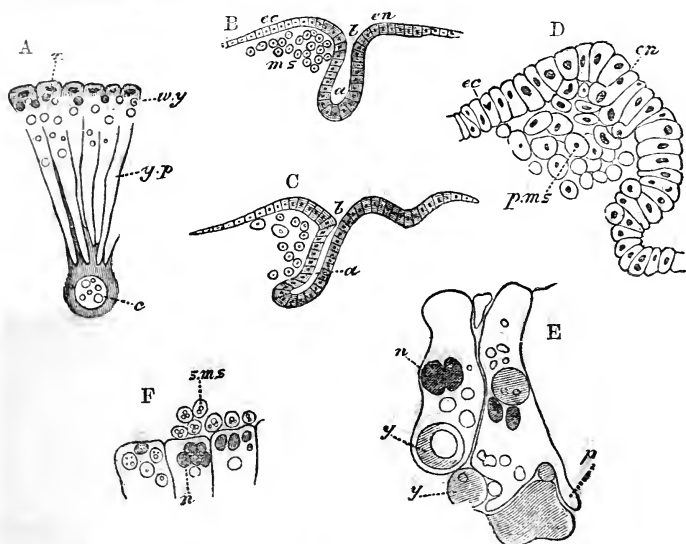
FIG 11.—*Figures illustrating the Development of Astacus.* (From Parker, after Reichenbach.) A. Section through part of the ovum during segmentation. *n.* nuclei; *w.y.* white yolk; *y.p.* yolk pyramids; *c.* central yolk mass. B and C. Longitudinal sections of the gastrula stage. *a.* archenteron; *b.* blastopore; *ms.* mesoblast; *ec.* epiblast; *en.* hypoblast, distinguished from epiblast by shading. D. Highly magnified view of anterior lip of blastopore, to show the origin of the primary mesoblast from the wall of the archenteron. *p.ms.* primary mesoblast; *ec.* epiblast; *en.* hypoblast. E. Two hypoblast cells to show the amœba-like absorption of yolk spheres. *y.* yolk; *n.* nucleus; *p.* pseudopodial process. F. Hypoblast cells giving rise endogenously to the secondary mesoblast (*s.ms.*); *n.* nucleus.

Nematelminthes.—Cucullanus. It grows backwards from the hypoblast cells at the persistent oral opening of the blastopore.

Tracheata.—Insecta. It grows inwards from the lips of the germinal groove (fig. 10), which probably represent the remains of a blastopore. Part of the mesoblast is probably also derived from the yolk-cells. A similar though more modified development of the mesoblast occurs in the Araneina (fig. 17).

Crustacea.—Decapoda. It partly grows in from the hypoblastic lips of the blastopore, and is partly derived from the yolk-cells (fig. 11).

2. The mesoblast is developed from the walls of hollow

outgrowths of the archenteron, the cavities of which become the body cavity.

Brachiopoda.—The walls of a pair of outgrowths form the whole of the mesoblast.

Chætognatha.—The mesoblast arises in the same manner as in the Brachiopoda (fig. 12).

Echinodermata.—The lining of the peritoneal cavity is developed from the walls of outgrowths of the archenteron, but the greater part of the mesoblast is derived from the amœboid cells budded off from the walls of the archenteron (fig. 13).

Enteropneusta (Balanoglossus).—The body cavity is derived from two pairs of alimentary diverticula, the walls of which give rise to the greater part of the mesoblast.
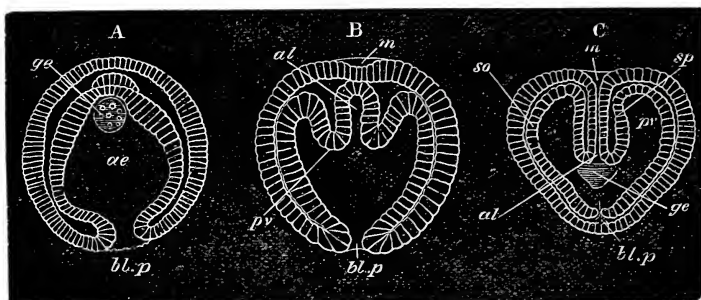
FIG. 12.—*Three Stages in the Development of Sagitta.* (A and C after Butschli, and B after Kowalevsky.) The three embryos are represented in the same positions. A. Represents the gastrula stage. B. Represents a succeeding stage, in which the primitive archenteron is commencing to be divided into three. C. Represents a later stage, in which the mouth involution (*m*) has become continuous with alimentary tract and the blastopore has become closed. *m.* mouth ; *al.* alimentary canal ; *ae.* archenteron , *bl.p*, blastopore ; *pv.* perivisceral cavity ; *sp.* splanchnic mesoblast ; *so.* somatic mesoblast ; *ge.* generative organs.

Chordata.—Paired archenteric outgrowths give rise to the whole mesoblast in Amphioxus (fig. 14), and the mode of formation of the mesoblast in other Chordata is probably secondarily derived from this.

3. The cells which will form the mesoblast become marked out very early, and cannot be regarded as definitely springing from either of the primary layers.

Turbellaria.—Leptoplana (fig. 15), Planaria polychroa (?)
Chætopoda.—Lumbricus, &c.
Discophora.

It is very possible that the cases quoted under this head ought more properly to belong to group 1.

4. The mesoblast cells are split off from the epiblast.

Nemertea.—Larva of Desor. The mesoblast is stated to be split off from the four invaginated discs.

5. The mesoblast is split off from the hypoblast.

Nemertea.—Some of the types without a metamorphosis.

Mollusca.—Scaphopoda. It is derived from the lateral and ventral cells of the hypoblast.

Gephyrea.—Phascolosoma.

Vertebrata.—In most of the higher Vertebrata the meso-blast is derived from the hypoblast (fig. 16).   In some types (*i. e.* most of the Icthyopsida) the mesoblast might be described as originating at the lips of the blastopore.
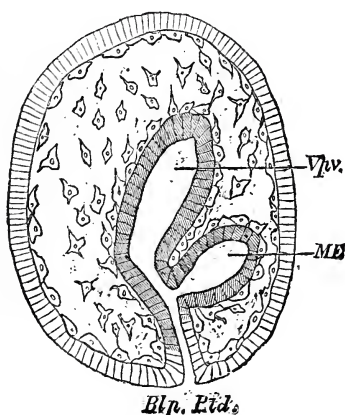
Fig. 13.—*Longitudinal Section through an Embryo of Cucumaria doliolum at the end of the fourth day*. *Vpv.* vaso-peritoneal vesicle : *ME.* mesenteron ; *Blp.*, *Ptd.* blastopore, proctodæum.

6. The mesoblast is derived from both germinal layers.

Tracheata.—Araneina (fig. 17). It is derived partly from cells split off from the epiblast and partly from the yolk-cells ; but it is probable that the statement that the mesoblast is derived from both the germinal layers is only formally accu-rate; and that the derivation of part of the mesoblast from the yolk-cells is not to be interpreted as a derivation from the hypoblast.

The conclusions to be drawn from the above summary are by no means such as might have been anticipated.   The analogy of the Cœlenterata would lead us to expect that the mesoblast would be derived partly from the epiblast and partly from the hypoblast.   Such, however, is not for the most part the case, though more complete investigations may

show that there are a greater number of instances in which
the mesoblast has a mixed origin than might be supposed
from the above summary.
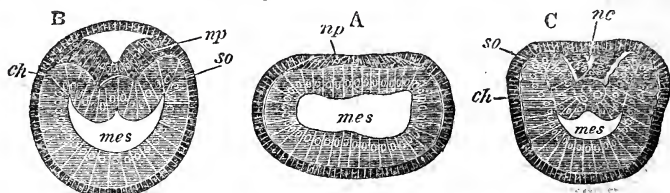
FIG. 14.—*Sections of an Amphioxus Embryo at three Stages.* (After Kow-
alevsky.)   A. Section at gastrula stage.   B. Section of a somewhat
older embryo.   C. Section through the anterior part of still older
embryo.   *np.* neural plate; *n.c.* neural canal; *mes.* archenteron in A,
and mesenteron in B and C; *ch.* notochord; *so.* mesoblastic somite.

I have attempted to reduce the types of development of
the mesoblast to six; but owing to the nature of the case it
is not always easy to distinguish the first of these from the
last four.   Of the six types the second will on most hands
be admitted to be the most remarkable.   The formation of
hollow outgrowths of the archenteron, the cavities of which
give rise to the body cavity, can only be explained on the
supposition that the body cavity of the types in which
such outgrowths occur are derived from diverticula cut off
from the alimentary tract.   The lining epithelium of the
diverticula—the peritoneal epithelium—is clearly part of the
primitive hypoblast, and this part of the mesoblast is clearly
hypoblastic in origin.
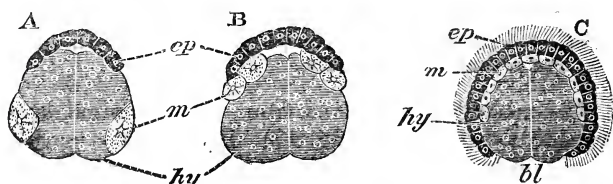
FIG. 15.—*Sections through the Ovum of Leptoplana tremellaris in three
Stages of Development.* (After Hallez.)   *ep.* epiblast; *m.* mesoblast;
hy. yolk cells (hypoblast); *bl.* blastopore.

In the case of the Chætognatha (Sagitta), Brachiopoda,
and Amphioxus, the whole of the mesoblast originates from
the walls of the diverticula; while in the Echinodermata the
walls of the diverticula only give rise to the vaso-peritoneal
epithelium, the remainder of the mesoblast being derived from
amœboid cells which spring from the walls of the archenteron

before the origin to the vaso-peritoneal outgrowths (figs. 2 and 13).

The first of these processes suggests the view that the whole of the mesoblast primitively arose by a process of histogenic differentiation from the walls of the archenteron. This view, which was originally put forward by myself (No. 4), appears at first sight very improbable, but it receives great support from the enormous development of the hypoblastic muscular system (Hertwigs, No. 13) in many Actinozoa. Lankester (No. 17), on the other hand, has urged that the mode of origin of the mesoblast in the Echinodermata is more primitive ; and that the amœboid cells which here give rise to the muscular and connective tissue represent cells which originally arose from the whole inner surface of the epiblast. It is, however, to be noted that even in the Echinodermata the amœboid cells arise from the *hypoblast*, and their mode of origin may, therefore, be used to support the view that the main part of the muscular system of higher types is derived from the primitive hypoblast.

Reserving for the moment the question as to what conclusions can be deduced from the above facts as to the origin of the mesoblast, it is important to determine how far the facts of embryology warrant us in supposing that in the whole of the triploblastic forms the body cavity originated from the alimentary diverticula. There can be but little doubt that the mode of origin of the mesoblast in the Vertebrata, as two solid plates split off from the hypoblast in which a cavity is secondarily developed, is an abbreviation of
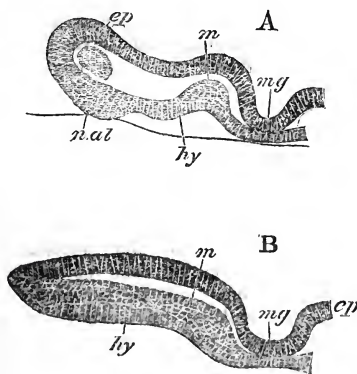
FIG. 16.—*Two Sections of a young Elasmobranch Embryo, to show the mesoblast split off as two lateral masses from the hypoblast. mg.* medullary groove; *ep.* epiblast; *m.* mesoblast; *hy.* hypoblast.

the process observable in Amphioxus; but this process approaches in many forms of Vertebrata to the ingrowth of the mesoblast from the lips of the blastopore.

It is, therefore, highly probable that the paired ingrowths of the mesoblast from the lips of the blastopore may have been in the first instance derived from a pair of archenteric diverticula. This process of formation of the mesoblast is, as may be seen by reference to the summary, the most frequent.[1]

While there is no difficulty in the view that the body cavity may have originated from a pair of enteric diverticula in the case of the forms where a body cavity is present, there is a considerable difficulty in holding this view, for forms in which there is no body cavity distinct from the alimentary diverticula.
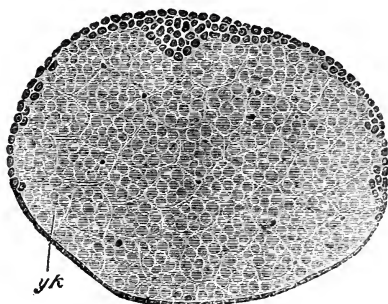
FIG. 17.—*Section through the Embryo of Agelena labyrinthica.* The section is represented with the ventral plate directed upwards. In the ventral plate is seen a keel-like thickening, which gives rise to the main mass of the mesoblast. *yk.* Yolk divided into large polygonal cells, in several of which are nuclei.

Of these types the Platyelminthes are the most striking. It is, no doubt, possible that a body cavity may have existed

[1] The wide occurrence of this process was first pointed out by Rabl. He holds, however, a peculiar modification of the gastræa theory, for which I must refer the reader to his paper (No. 23); according to this theory the mesoblast has sprung from a zone of cells of the blastosphere, at the junction between the cells which will be invaginated and the epiblast cells. In the bilateral blastosphere, from which he holds that all the higher forms (bilateralia) have originated, these cells had a bilateral arrangement, and thus the bilateral origin of the mesoblast is explained. The origin of the mesoblast from the lips of the blastopore is explained by the position of its mother-cells in the blastosphere. It need scarcely be said that the views already put forward as to the probable mode of origin of the mesoblast, founded on the analogy of the Cœlenterata, are quite incompatible with Rabl's theories.

in the Platyelminthes, and become lost, and even in some cases replaced physiologically by alimentary diverticula. The usual view of the primitive character of the Platyelminthes, which has much to support it, is, however, opposed to the idea that the body cavity has disappeared.

If Kowalewsky[1] is right in stating that he has found a form intermediate between the Cœlenterata and the Platyelminthes, there will be strong grounds for holding that the Platyelminthes are, like the Cœlenterata, forms the ancestors of which were never provided with a body cavity.

Perhaps the triploblastica are composed of two groups, viz. (1) a more ancestral group (the Platyelminthes), in which there is no body cavity as distinct from the alimentary, and (2) a group descended from these, in which two of the alimentary diverticula have become separated from the alimentary tract to form a body cavity (remaining triploblastica). However this may be, the above considerations are sufficient to show how much there is that is still obscure with reference even to the body cavity.

If embryology gives no certain sound as to the questions just raised with reference to the body cavity, still less is it to be hoped that the remaining questions with reference to the origin of the mesoblast can be satisfactorily answered. It is clear, in the first place, from an inspection of the summary given above, that the process of development of the mesoblast is, in all the higher forms, very much abbreviated and modified. Not only is its differentiation relatively deferred, but it does not in most cases originate, as it must have done to start with, as a more or less continuous sheet, split off from one or both the primary layers. It originates in most cases from the hypoblast, and although the considerations already urged preclude us from laying very great stress on this mode of origin ; yet, as suggested above, it appears to me not impossible, judging from the analogy of the Actinozoa, that the muscular system of the triploblastica may have primitively mainly arisen from differentiations of the hypoblast of the alimentary diverticula, which seem to have given rise to the body cavity.

The great changes which have taken place in the development of the mesoblast would be more intelligible on this view than on the view of the mesoblast having primitively largely originated from the epiblast. The presence of food-

[1] 'Zoologischer Anzeiger,' No. 52, p. 140. This form has been named by Kowalewsky *Coleoplana Metschnikowii.* Kowalewsky's description appears, however, to be quite compatible with the view that this form is a creeping Ctenophor, in no way related to the Turbellarians.

yolk is much more frequent in the hypoblast than in the epiblast; and it is well known that a large number of the changes in early development are caused by food-yolk. If, therefore, the mesoblast has been derived from the hypoblast, many more changes might be expected to have been introduced into its early development than if it had been derived from the epiblast. At the same time the hypoblastic origin of the mesoblast would assist in explaining how it has come about that the development of the nervous system is almost always much less modified than that of the mesoblast, and that the nervous system is not, as might, on the grounds of analogy, have been anticipated, developed in the mesoblast.

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(15) A. KOWALEWSKY. "Embryologische Studien an Warmern u. Arthropoden," 'Mem. Acad., Petersbourg,' series vii, vol. xvi, 1871.

(16) E. R. LANKESTER. "On the Germinal Layers of the Embryo as the Basis of the Genealogical Classification of Animals," 'Ann. and Mag. of Nat. Hist.,' 1873.

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HUBRECHT'S RESEARCHES *on the* NERVOUS SYSTEM *of* NEMERTINES.[1] (With Plate XXIII).

THE above cited paper, which has just been published by the Amsterdam Academy of Sciences, and from which we have copied figures 1 to 12 on Plate XXIII, gives a detailed account of the situation and the structure of the central nervous system in the Nemerteans.   After all that has been said on this subject in the works and treatises of *McIntosh*, *Quatrefages*, *Keferstein*, and in a former paper of the same author, it would seem superfluous once more to go over the same ground.   Only it should be here remembered that the histological structure was not, or very insufficiently, considered by those authors, whereas, in the present paper, due space is allowed to the description of the histological details.   The minute structure of the so-called " side-organs " (McIntosh's cephalic sacs) is here also for the first time minutely entered into.

A series of experiments is next recorded in favour of the author's hypothesis, that in the Lineidæ these "cephalic sacs" with their internal ciliated canal and with the deep longitudinal slits on each side of the head must be regarded as a special apparatus serving for respiratory purposes, the oxygen being taken hold of by the hæmoglobin contained in the nerve-cells belonging to the central nervous apparatus as it stretches throughout the whole length of the animal, from the head to the tip of the tail.   Other arguments in favour of this hypothesis are given, some of them derived from the phylogenetic, others from the ontogenetic development of these organs.

Finally, the paper discusses an explanation of the origin of the dorsal nerve-cord of Vertebrates and the ventral nerve-cord of Arthropods and Annelids out of originally paired lateral cords.   This explanation is suggested by the situation of the latter in different genera of Nemerteans, and finds itself in harmony with general views which were already expressed on former occasions by Gegenbaur, Harting, Balfour, and others.   Two hitherto unknown facts resulting from the author's investigations are more specially brought to bear upon this point; 1st, the presence in all *Hoplonemertini* as yet examined on this head, of a commissure uniting the two lateral nerve-cords and situated *above* the intestine

immediately before the anus (*Moseley* describes such a commissure in *Pelagonemertes*); and 2nd, the displacement of the lateral nerve-cords in the author's new genus *Langia*, which tend to approach each other *on the dorsal side* (not ventrally as they do in the genera *Drepanophorus* and *Oerstedia*), at least in the anterior portion of the body. This explanation would restore the homology between the dorsum of Vertebrates on the one hand and of Arthropods and Annelids on the other, a homology which of late years has been put into serious doubt by the researches of *Dohrn* and *Semper*, whose ingenious suggestions have as yet, however, never definitely overcome certain primary objections inherent to their views, which correspond as a whole with Geoffroy St. Hilaire's saying that insects are Vertebrates walking on their backs.

After this rapid exposition of the contents of the paper, we shall give a somewhat more detailed account of certain parts of it. It should here be remarked that the nomenclature of the genera and suborders employed is that proposed by the author in a former paper,[1] in which he divides the Nemerteans into three suborders: PALÆONEMERTINI, with the genera *Carinella*, *Cephalothrix*, *Polia* and *Valencinia;* SCHIZONEMERTINI, with the genera *Lineus*, *Borlasia*, *Cerebratulus* and *Langia;* HOPLONEMERTINI, with the genera *Amphiporus*, *Drepanophorus*, *Tetrastemma*, *Oerstedia*, *Prosorhochmus*, and *Nemertes*.

A. *The central nervous system.*—As such the author does not regard the cephalic ganglia only—as was done by all his predecessors—but the so-called longitudinal nerves as well, on account of the sheath of ganglion cells which uninterruptedly accompanies these trunks from their origin in the cephalic lobes down to the extremity of the tail, in all the genera without exception.

In the genus *Carinella*—which appears to be one of the more primitive and less differentiated—the whole central nervous system is situated immediately under the epidermal tissues, outside the muscular body wall (Pl. XXIII, fig. 7), and the cephalic ganglion takes the form of a simple anterior enlargement of the lateral trunks. No distinct division into lobes can be detected in transverse sections; the ventral commissure is very broad, the dorsal commissure exceedingly thin; through the ring thus formed the proboscis and its sheath passes (Pl. XXIII, fig. 2). The mouth opens behind and under the ganglion. The fibrous nerve-sub-

[1] The "Genera of European Nemerteans Critically Revised," 'Notes from the Leyden Museum,' vol. i, p. 193.

stance prevails over the cellular in this genus, and is some-what loosely arranged. It is *not* surrounded by nerve-cells, as these form only an *external* coating to it, whereas in the higher-developed genera the cellular substance (in the brain at least, not always in the lateral trunks) *does* surround it on all sides (Pl. XXIII, figs. 4, 5, and 6). This cellular portion in *Carinella* is also of a less compact nature than in those of more differentiated genera, and is everywhere in direct contact with the epidermoidal tissue. This may be regarded as the more primitive stage, in which the entire central nervous system, from the head of the animal down to the tail, has not yet become separated from the ectoderm through intervening muscular tissue.

The SCHIZONEMERTINI and the genera *Polia* and *Valencinia*, among the PALÆONEMERTINI, represent a further stage of development, inasmuch as the central nervous apparatus is here situated exteriorly to the circular and interiorly to the outer longitudinal muscular layer, and so is everywhere surrounded by muscular tissue; whereas in the large majority of these genera the longitudinal trunks ("Nervenmarkstämme," as the author proposes to designate them) occupy a strictly lateral, opposite position (Pl. XXIII, fig. 8) the genus *Langia*, with the curious longitudinal depression along the back, shows the well-marked tendency, already mentioned above, of an approximation of these two parallel trunks on the dorsal side (Pl. XXIII, fig. 9). In none of all these genera could a commissure uniting the lateral trunks in the posterior extremity of the animal be detected. They, simply terminate. This commissure makes its appearance in the HOPLONEMERTINI, which are, moreover, characterised by the lateral trunks being situated wholly *interiorly* to the muscular body wall. So in this respect they form the opposite extreme to *Carinella*.

Generally the position of the nerve-trunks is strictly lateral (Pl. XXIII, fig. 10), also in this suborder; in certain genera, however, as *Drepanophorus* (Pl. XXIII, fig. 11), the longitudinal trunks have approached each other on the ventral side, and would, indeed, if they were connected by commissures or eventually coalesced, form an œsophageal nerve-ring and a ventral cord corresponding to the "Bauchmark" of Annelids and Arthropods. But such commissures fail in this genus, as they do in all other Nemerteans, and even in *Drepanophorus* the longitudinal trunks bend upwards in the extremity of the tail, and are in direct communication by a commissure *above* the digestive tract. This large amount of variability in the position of the central

nervous apparatus, with respect to the muscular layers of the body wall on the one hand, and to the axis of the body on the other hand, seems, indeed, to justify the author's conclusion that the *Nemertines* represent an ancient and primitive stock in which an apparatus, otherwise so stable as the central nerve chain, is subject to interesting variations, which point to higher stages of differentiation, apparently wide apart, which, however, are reached in other subdivisions of the animal kingdom.

B. *The side organs.*—The curious apparatus, accessory to the Nemertean brain, which has been designated by so many diverse names (side organs, cephalic sacs, ciliated furrow, cephalic grooves, &c.), is here for the first time studied in the light of comparison. The author, who has had at his disposal all the different genera of European Nemerteans, has been able to study this apparatus in the most various stages, and develops his views as to its probable phylogenetic development, basing his views in the first place on the comparative anatomy, and secondly, on certain facts which recent embryological observations have brought to light.

The lowest stage of differentiation of this apparatus is present in *Carinella annulata* (we leave *Cephalothrix,* where it seems to be altogether absent, out of consideration), in which genus the epiderm shows a transverse, shallow furrow, which is interrupted in the median line of the back, slightly bent, and situated about in a level with the middle of the brain. This groove is ciliated. Series of transverse and horizontal sections showed no traces of any complication of this arrangement (Pl. XXIII, fig. 2,). Where this furrow is situated, the distance separating the nerve-cells from the external ciliated surface of the body—a distance which all along the nerve-trunks is very insignificant indeed —is still more reduced, and the facility for osmotic intercourse augmented.

Carinella inexpectata, a new species which the author has elsewhere described,[1] agrees with *Carinella annulata* in all general respects. It differs from it in showing the apparatus just described more complicated by a few steps. Instead of the simple exterior transverse groove this species carries short parallel grooves placed perpendicular with respect to the one larger transverse one and confluent with it. In the middle of this transverse groove, strictly laterally, there is a small opening leading into a ciliated duct. This duct penetrates the ectodermic tissues, and immediately enters amidst

[1] 'Notes from the Leyden Museum, vol. i, p. 93.

the ganglion cells of the brain.   It there appears to terminate
blindly (Pl. XXIII, fig. 3.)   In all other respects the structure
of the brain and nervous system closely agrees with *C. annu-
lata.*

A further degree of complication is attained in *Polia,*
where the brain no longer lies immediately under the epi-
derm, but is separated from it by intervening muscular tissue.
Exteriorly the ciliated transverse grooves, with numerous
small transverse furrows perpendicular to it, have very much
the appearance of the grooves in *Carinella inexpectata.*

Sections show that here the ciliated duct traverses the
muscular tissue as well, enters between the nerve-cells of the
brain, at the same time making a double bend (Pl. XXIII,
fig. 4), and finally terminates in what appears to be a
more or less swollen enlargement.   Another complication
has taken place, in so far as the brain is no longer a simple
enlargement of the lateral trunks, but is split up into lobes,
two dorsal ones and two ventral ones.   Moreover, the dorsal
lobes are separated into a larger anterior and a smaller pos-
terior portion, applied to the former as a sort of cap.   That
such a separation exists is rarely visible by transparency; it
can only be clearly made out in horizontal sections.   Only
into this third portion of the brain, in this posterior dorsal
lobe, the ciliated canal communicating with the exterior
penetrates.   In its turn this posterior cerebral lobe is capped
over postero-medially by a layer of larger cells with distinct
nuclei, which in their appearance strongly resemble the cells
belonging to the coating of the œsophagus, which cells lie
in their immediate vicinity in the same section.   The situa-
tion of this layer of cells is indicated by a paler tint in
Pl. XXIII, fig. 4.   Further down we shall mention the
author's views respecting the morphological significance of
this group of cells.

In all the SCHIZONEMERTINI the external appearance of
the apparatus is considerably different, in so far as there is
no more any transverse furrrow or small opening of the
ciliated canal, but two deep longitudinal slits on each side
of the head, situated between the mouth and the tip of the
snout.   These slits penetrate deeply into the muscular
tissue of the head, and in the bottom of them the lobes of
the ganglion stand out more or less freely, only protected
by a very thin layer of tissue.   Their internal surface is
covered by numerous and long cilia, and in their postero-
medial portion they are continued into the ciliated canal
which here penetrates into the third (posterior) ganglionic
lobe.   The author regards the cephalic slits in this suborder

(to which the majority of hitherto known Nemerteans belong) as entirely homologous with the simple opening described for *Polia, Valencinia, &c.;* this opening having here been widened out and deepened for purposes in direct connection with the strongly augmented amount of hæmoglobin contained in the nerve-cells, and with the scarcity of oxygen in those places where the species of this suborder are found (deep in the mud, under stones, or in decaying matters on the sea bottom). The respiratory function of these slits, which was tested by the author in different physiological experiments, described in the original memoir, can be exercised, 1st, at the surface of the two pairs of anterior lobes, where these are externally visible at the bottom of the slits, and 2nd, in a more complete way inside the third (posterior) cerebral lobe (which has here become somewhat more separated from the anterior than was the case in *Polia*), where the ciliated duct with its continual current of fresh seawater is immediately surrounded by the hæmoglobinous nerve-cells (see Pl. XXIII, fig. 5). In some of these species the ciliated canal divides into two, one penetrating among the nerve-cells, the other apparently remaining more externally situated in relation to the ganglionic lobe. Both have blind terminations, and never show any trace of a special sensory epithelial layer. The accessory mass of large cells (pale in fig. 5) is somewhat more conspicuous but occupies the same place as in *Polia*.

In the HOPLONEMERTINI differentiation has gone on along another tract. The transverse furrow has not been modified into a longitudinal slit, but in certain genera (*Drepanophorus, Amphiporus*) it is even provided with short longitudinal grooves perpendicular to it, exactly as it is found in *Polia*, whereas in other genera these secondary short grooves are absent. Internally, another important change is the entire separation of the posterior cerebral lobe from the anterior dorsal one, with which it is still so closely united in *Polia*, and with which it is here only connected by one or more nervous commissures. With this separation a greater independence in its position has been gained : it may either be situated behind, on a level with, or before the rest of the brain. Here, as in *Polia* and the SCHIZONEMERTINI, there is a group of cells of quite different appearance coalesced with the ganglion cells, which latter, however, form the bulk of the posterior lobe ; but here a ciliated space or canal remains free in the midst of this accessory mass of cells. The primary ciliated canal penetrates from the exterior into the midst of the true ganglion cells ; into

it the second ciliated canal above mentioned opens, as indicated in the figure (Pl. XXIII, fig. 6).

Summarising the different phases in which this accessory apparatus to the brain is found in the various forms of Nemerteans, we have—*a*. A simple transverse furrow in the epiderm, on a level with the brain, which lies immediately under the epiderm. *b*. A slight complication of the furrow, and an external opening from which a short canal leads into the mass of nerve-cells from the brain (which in position and structure has undergone no change). *c*. A separation of the brain into three lobes, into the posterior one of which the ciliated canal, perforating the muscles and opening exteriorly, penetrates, whilst a mass of cells strongly resembling the cells of the wall of the œsophagus coalesces with this posterior lobe. At the same time we have sub. *c*, the three following modifications :—*a*. The external furrow as in *b*, the internal canal simply bent. β. The external opening modified into a deep longitudinal slit on both sides; the internal canal either simply bent or divided into two. γ. The external furrow as in *b* and *c a*; the ciliated canal divides into two, one branch situated amidst the true ganglion cells, the other in the accessory mass of cells.

As to the development of the apparatus, it has as yet only been studied in a few species. From the results arrived at by *Metschnikoff, Leuckart, Pagenstecher, Bütschli,* and *Barrois,* the author feels justified in concluding that—1st. The exterior opening and ciliated canal leading amongst the ganglion cells of the brain are an invagination from the ectoderm. 2nd. The mass of larger cells (indicated by lighter shading in the figs.), in which sometimes a ciliated space persists, is an outgrowth from the œsophagus, afterwards separated from it; by coalescence with the invagination of the ectoderm the internal space sometimes communicates with the exterior. 3rd. The bulk of the apparatus is directly derived from the brain, with which it remains in more or less intimate connection.

The facts mentioned under the second heading would be a strong argument in favour of this apparatus having a respiratory function. The gills of Balonoglossus develop in the same way; whereas here it is the blood fluid with. which similar outgrowths of the œsophagus become more intimately connected, it is in Nemerteans the hæmoglobinous nerve-tissue.

The author is not unwilling to recognise that in the higher differentiated HOPLONEMERTINI the apparatus may gradually lose its significance as a special respiratory centre,

especially because here the circulation is more perfected and
the blood-corpuscles are hæmoglobinous themselves, the
nerve-tissue, at the same time, losing its deep-red tinge.
Only he adds that, not being able to detect any sensory epi-
thelium in these organs, he is provisionally inclined to doubt
whether, after having lost their primary respiratory signifi-
cance, they ever become secondarily modified into a sense-
organ in the higher differentiated forms.

c. *Histology of the nerve-centres, peripheral nerves, &c.*
—Respecting the further histological and anatomical results,
we may rapidly note the following :—The ganglion cells in
the brain, in all genera above *Carinella* and *Cephalotrix*,
surround the central mass of nerve-fibre on all sides. This
fibrous " skeleton" of the central apparatus is figured for the
SCHIZONEMERTINI in Pl. XXIII, fig. 1 ; the right dorsal
lobe is supposed to have been removed in order to show the
ventral one. The fibrous nerve-substance is indicated by a
paler, the cellular by a darker tint. The general shape and
distribution of this fibrous nucleus of the brain may be
gathered from the figure ; it is also continued into the posterior
brain lobe.

A very thin homogeneous layer separates the fibrous from
the cellular nerve-substance ; the processes of the nerve-
cells (very few multipolar, the majority *apparently* unipolar)
are seen to perforate this homogeneous layer, and to pene-
trate into the central fibrous substance. The larger nerves
for the eyes and the tip of the snout, those for the proboscis,
and those for the œsophagus, are seen, as well on compression
as in sections, to take their origin in this central fibrous
nerve-skeleton. The two latter sets of nerves are for the
first time correctly described by the author. For *Carinella*
he succeeded in demonstrating the direct passage of strong
nerves, leaving the brain close to the ventral commissure
into the proboscis, where they continue their course longi-
tudinally between the muscular and the cellular coating.
Identical nerves were clearly made out in certain SCHIZO-
NEMERTINI, and for some of the HOPLONEMERTINI *von
Kennel's* suggestion that *McIntosh's* " beaded layer" in the
proboscis of this group is of a nervous nature was fully con-
firmed, and at the same time the distribution of delicate and
numerous nerve-twigs going to the papillæ of this proboscis
was made out.

Next to this proboscidian nerve another strong nerve leaves
the brain, springing from the posterior portion of the inferior
lobes. This pair of nerves is present in all Nemerteans, and
is intended for the anterior portion of the alimentary canal.

on the surface of which it is subject to further dichotomic division. To this nerve the author assigns the name of *N. vagus,* a term already employed by Leydig for other Invertebrates, and recently misapplied by Semper (who was then unacquainted with the presence in Nemerteans of a nerve such as the one here mentioned) to the dorsal commissure of the cerebral ganglia in Nemertines.

A thin longitudinal nerve, originating from the dorsal commissure and situated medially and dorsally, is looked upon by the author as a special nerve for the proboscidian chamber (Pl. XXIII, fig. 1).

The anal commissure, situated *above* the intestine mentioned above, presents no further particulars, its high morphological interest depending on the fact of its causing the whole of the paired symmetrical central nervous system present throughout the whole length of the animal to be situated, together with all the three transverse commissures, *above the alimentary canal.*

The system of peripheric nerves is not exhaustively treated of in the present paper ; the author's investigations on this point being not yet terminated. Still, certain points which may eventually prove to be interesting are already noticed. Whereas in the HOPLONEMERTINI where the lateral nerve-trunks are situated inside of the muscular body wall, small pairs of nerves springing from those trunks placed metamerically, and distributing themselves to the muscular coats of the body wall can easily be detected, no such transverse stems to the longitudinal trunks could be detected in the SCHIZONEMERTINI. *Only here the author found a cellular layer, which he holds to be a nervous layer,* encircling the body in the way of a tunic, and situated in a free space between the outer longitudinal and the circular muscular layers (Pl. XXIII, fig. 12,). The cells of this layer are multipolar, connected together as a sort of network, and uninterruptedly passing into the ganglion cells ensheathing the longitudinal trunks. Further details on the structure and significance of this layer are postponed by the author to a future paper on this subject.

On the STRUCTURE *of the* NEPHRIDIA *of the* MEDICINAL
   LEECH. By A. G. BOURNE, Assistant in the Zoological
   Laboratory of University College, London. (With
   Plates XXIV and XXV.)

THE investigation into the structure of the so-called seg-
mental organs, or (better) " nephridia " of the medicinal
Leech, of which the following pages give an account, was
undertaken at the suggestion of Professor Lankester and
carried out under his direction and with his supervision and
advice in the zoological laboratory of University College,
London.[1]

A series of very successfully prepared sections of the
Leech, made by Mr. J. E. Blomfield, of Oxford, at present
Demonstrator in Prof. Lankester's laboratory, had sufficed
to draw our attention to a number of interesting points in
the minute structure of that animal.

Amongst others, Professor Lankester especially pointed
out the remarkable structure of the constituent cells of the
nephridia, each cell being either perforated by a simple duct
or having within its substance an arborescent extension of
the system of ducts, common to the whole gland. Further,
each cell was seen to be enclosed in a mesh of the capillary
system filled with red fluid (the hæmal system), which is
spread throughout the tissues of the medicinal Leech.

I undertook the more thorough investigation of these
structures with two objects in view. In the first place, since
the community of origin of the renal organ of Vertebrates,
Worms, and Molluscs, has become a well-established theory,
it cannot but be a matter of the first importance that the
complete anatomy of the various modifications of the typical
" NEPHRIDIUM " (as Professor Lankester[2] has termed the
single excretory tube from the multiplication or branching
of which more complex forms, such as the " primitive kid-
ney " or archi-NEPHRON, have been derived) should be care-
fully worked out.

In the second place, it is known that in Leeches this
organ presents very wide differences of structure in different
genera, differences which appear to go hand in hand with
corresponding modifications of the hæmal system, and I was
therefore anxious to obtain a complete knowledge of the

[1] See his " Observations on the Microscopic Anatomy of the Leech," in
the 'Zoolog. Anzeiger,' 1880, No. 49.
[2] " Notes on Embryology and Classification," this Journal, 1877.

arrangements obtaining in *Hirudo medicinalis*, in order subsequently to compare with the nephridia of that form— those of Nephelis, Clepsine, and Pontobdella.

I.—CURRENT STATEMENTS AS TO THE NEPHRIDIA OF THE MEDICINAL LEECH.

Omitting detailed reference to the writings of De Blainville, to Brandt and Ratzeburg's " Medizinische Zoologie," and to Moquin Tandon's valuable "Monographie de la famille des Hirudinées." I shall here give brief extracts from Gratiolet, from Leuckart, from Leydig, and from Gegenbaur, in order to show what is the present state of our knowledge as to the nephridia of the Leech.

In 1850 Gratiolet published a memoir in the French Academy's series on the " Circulation of the Medicinal Leech," and in 1862 another paper on this subject (" Ann. des Sciences Nat.," Ser. iv, ' Zool.,' 1862).

Gratiolet says :—" The segmental organs, as Dr. Williams has called them, have been considered as *appareils aquifères* (? water vessels), as tracheal or respiratory organs, or as mucous glands ; further, Dr. Williams (*loc. cit.*) has suggested that these segmental organs are the true ovaries in all the annelids.   They are seventeen in number on each side of the body.   Each is constituted by—1. A tube with thick glandular walls.   2. A vesicle behind the tube, opening on the ventral surface of the animal by an excretory orifice.   The tube is bent up into a narrow loop, the much attenuated branches of which end by anastomosing; it is spread into a ' sling,' and then bent back into a right angle, one extremity of the ' sling ' being nearly vertical, the other directed horizontally forwards, and ending by being rolled on itself in a sort of bud, the first and much the thickest end being more or less dilated according to its position."   Gratiolet means by this that the two halves of the lobe folded on itself, which forms the vertical portion, may lie close together, or be separated by an interval.

He continues :—" The two branches of the loop are similar in structure, the anterior differing from the posterior in having a canal at the lower end of the vertical part, which opens on the upper wall of the well-known oval or spherical vesicle.

" In front of the series of testes the upper parts of the loops are much dilated ; the lower part bends forwards, and end in the above-mentioned bud ; to this in the segments containing the testes is added a small cœcal process, which

leads inwards toward the middle line; these processes end blindly on the upper surface of the testes. The two nephridia behind the last testis also possess these cæca, which are directed inwards and end in the same way.

" This connection with the testes is very interesting, as it reproduces in a very significant manner the relations which certain analogous organs have with the testes in the Oligochæta."

Gratiolet continues:—"Various erroneous views have been entertained with regard to the vesicles; they have been thought to be tracheæ or spiracles by Schlacht and Bibiena, and Thomas, Dugès, and Audouin have considered them as lungs. Dugès has thought that the glandular part might be a sort of heart.

" It is not absolutely contrary to probability to consider them as water-vessels, but how they could subserve aerial respiration is difficult to explain.

" Nevertheless, these hypotheses, however rash they may have been, are nothing to that which has recently been put forward and defended by Dr. Williams.

" According to this author, the glandular parts are the normal ovaries. The eggs develope and ripen in their branches. Each complete segment of the body has thus its female genital organ—its ovary."

Gratiolet agrees with De Blainville, Brandt, and Moquin Tandon that these organs are secretory—are, in fact, renal organs. D'Udekem has also shown this to be the case.

Gratiolet considers that they also serve to keep the skin moist while the animal is out of the water, and correlates the greater power the medicinal Leech has of staying out of water compared with that of the Horse Leech with the larger size of these organs in the former animals. Leydig has shown, however, that unicellular glands open all over the surface of the skin, and these would serve to keep it moist, just as in land Planarians, the frog, and other terrestrial animals which possess a moist skin.

I see no reason to suppose that the nephridia of the Leech have any such mucous function.

Leuckart ('Die Menschlichen Parasiten,' vol. i, 1863) has given a brief description of the nephridia. The most important new fact he mentions is that the vesicle has considerable contractile power, which is produced by a delicate muscle which lies outside the *tunica propria*. The inner surface of this membrane bears a finely granular parchment epithelium composed of large cells.

Leuckart also states that there are muscle-fibres in the

walls of the central duct. This I have not found to be the case.

Leydig (' Histologie des Menschen und der Thiere ') added to previous knowledge of the general form of the nephridia of the Leeches (which was largely due to his own researches, especially that entitled " Zur Anatomie von Piscicola," ' Zeitsch. wiss. Zoologie,' vol. i), an important fact of minute structure in which it is now known to agree with the ne- phridia of some other types, such as earthworm and larval Pulmonate Gastropods. He showed, in fact, by a figure to which there is no further allusion in the text, that the gland-cells of this organ are perforated by ducts, so that the cells have the form of hollow cylinders ; but he did not observe the more complicated condition of arborescent ducts discovered by Lankester, which I shall describe hereafter. The perforated cells figured by Leydig in his ' Histologie ' were from the nephridium of Hæmopis. Claparède first showed that such was also the relation of duct and cell in the nephridium of the common earthworm (" Histologie des Regenwurms," ' Zeitsch. wiss. Zool.,' vol. 1869).

Gegenbaur was the first to point out the complicated labyrinthine character of the ducts of the nephridium of the Leech when comparing that organ with the nephridium of the earthworm (' Zeitsch. wiss. Zool., vol. iv), which he was the first to describe accurately. The condition of knowledge at the time when Gegenbaur's observations were pub- lished (five-and-twenty years ago) did not require a minute histological account of these organs, and, accordingly, we do not find histological details in his memoir. Excepting for the figure of cells from the nephridium of Hæmopis, pub- lished by Leydig, there appears to have been no attempt to inquire further into the structure of this organ during later years ; and we have the following account in Gegenbaur's ' Elements of Comparative Anatomy ' (English edition, 1879), which may be taken as representing the actual state of knowledge with regard to them. It will be observed as an important point distinctive of the nephridia of the genus Hirudo that no internal opening has been observed leading from the body-cavity into the duct of the nephridium, and to the possible existence or non-existence of such an opening my observations have, *inter alia*, been carefully directed.

Gegenbaur says (p. 176) :—" So far as the structure of the excretory organs is concerned, few fresh characters appear in the Annulata (Hirudinea, Oligochæta, Chætopoda). The organs correspond to the metamerism of the body, for they are regularly distributed on either side of almost every one

of its segments. They have, therefore, though with but little reason, been called ' segmental organs,' a name which is just as suitable for many other organs. Each of them consists of a closely-coiled or loop-like canal (Schleifen canale), which has an internal opening, often peculiar in form and always ciliated, and which opens at the other end on to the surface of the body. This canal is sometimes similar in character throughout its whole length, or but slightly differentiated ; frequently several segments may be made out in it, which generally correspond with those already described in the Platyhelminthes and Rotatoria. The innermost portion, on which the opening into the cœlom is placed, is ordinarily the longest; it is distinguished by its funnellike or rosette-shaped orifice. In the next portion the walls may be seen to be glandular in structure. The last portion, which is frequently widened, is provided with a layer of muscle ; it almost always opens on to the ventral surface. These organs are no more purely excretory in function than they are in other Vermes, for we not unfrequently find them entrusted with other functions.

" In the Hirudinea these organs are preceded, in the embryonic stage, by three pairs of looped canals, which are not connected with those formed at a later stage ; they are found in the posterior half of the ventral surface. In structure they are similar to, but simpler than, the permanent canals, and disappear after these are developed. This most important fact shows that the looped canals of the Annulata cannot be regarded as the direct homologues of the excretory organs of the lower Vermes; at the same time arises the question, Are the looped canals of those Annulata, which show no signs of primitive organs of this kind, comparable to the permanent looped canals of the Hirudinea, or only to the primitive ones ?

" In their more special characters there is great variety among the Hirudinea, the canals in one division having no internal opening. Instead of this they begin with a closed portion in the form of a loop, which consists of numerous canals united with one another into a labyrinth (*Hirudo*). From these looped organs a single canal is given off which opens by a vesicular enlargement on the surface of the body. In others (*Clepsine, Nephelis*) the labyrinthine portion is present, but it has an internal opening, which projects into the lateral blood-sinuses of the body."

I may be allowed further to cite a statement of Mr. Balfour's relative to the nephridium of the Leech, which will serve to illustrate the view generally entertained as to its

structure. After describing (this Journal, 1879, p. 432) the segmental organs (nephridia) of the interesting Quatho-pod, Peripatus, as consisting of 1, a dilated vesicle; 2, a coiled glandular tube; 3, a short terminal portion, he proceeds to say that though formed on a type of their own, the segmental organs of Peripatus more closely resembles those of the Leech than of any other form. The facts of the structure which I have to record as the result of my studies do not allow me to accept the view that there is any special similarity between the simple nephridium of Peripatus and the complicated one of Hirudo. The high differentiation of the nephridium of Hirudo appears in fact to have been overlooked by zoologists generally.

II.—METHODS OF INVESTIGATION.

1. For the purpose of studying the nephridium entire and in the fresh state, Leeches were chloroformed and pinned in a gutta-percha trough, and opened by a dorsal median incision, the trough being filled with normal salt-solution. In this way any one of the series of seventeen pairs of nephridia could be dissected out whole and removed to a glass slide for observation under the microscope.

The transparency of the organ renders this method of study of great value.

2. In order to obtain permanent and stained preparations, the nephridia, after excision, were placed in $\frac{1}{2}$ per cent. osmic acid for twenty minutes, washed in normal salt solution or dilute alcohol and then stained for half-an-hour with Ranvier's picrocarmin. Finally, they were mounted in glycerine.

Other whole preparations of great value in the difficult task of following out the course of the ducts and ductules, were obtained by placing whole nephridia in absolute alcohol, staining with hæmatoxylin, clarifying with clove-oil and mounting in Canada balsam.

3. Teazed preparations for the purpose of studying the minute characters of the tissues composing the organ were used. The nephridia were teazed either fresh, in salt solution or in osmic acid, or after short maceration (twenty minutes) in nitric acid (20 per cent. in water), or in Weissmann's potash solution (40 parts caustic potash to 100 parts water), or after prolonged maceration (six months) in 2 per cent. aqueous solution of potassium bichromate. The latter method yielded valuable results, as also did maceration in $\frac{1}{10}$ per cent. chromic acid.

4. The method of silver-staining was also applied to fresh

nephridia in order to ascertain the structure of the walls
of the blood-vessels and of the ducts of the gland. An
interesting structure in the substance of the gland-cells
themselves was revealed by this method.

5. Injection of the vascular system with soluble Berlin-
blue was practised with some success so far as the injection of
the finer vessels of the integument, &c., was concerned, but I
have not succeeded in injecting the capillaries of the nephri-
dium itself. I have been at present equally unsuccessful in
injecting the ducts of the nephridium. The mode of injection
made use of was that used by Moseley in his injections of in-
sects, which consists in using a small glass tube drawn out
to a capillary termination serving as the nozzle, a caoutchouc
tube being attached to the other end of the glass tube. When
the little apparatus is filled with injecting fluid it suffices to
compress the caoutchouc tube in order to obtain sufficient
pressure and sufficient flow of liquid for the injection of very
minute organs. I also made with Professor Lankester some
injections of indigo carmine into the body substance of an
uninjured Leech by means of a subcutaneous syringe. The
Leeches lived well after receiving a cubic centimètre of the
indigo-carmine solution, but we have not as yet obtained
any definite results as to the excretion of indigo by the
nephridia or other organs.

6. The most important method of which I availed myself
in conjunction with the study of whole nephridia, is that
of section-cutting; sections of nephridia were obtained either
by cutting whole Leeches or by cutting only a single nephri-
dium and its surrounding tissues isolated for the purpose.

In either case the Leech was prepared by killing with
chloroform, and was *moderately* stretched by means of a pin
at each end fixing it in a gutta-percha trough. Chromic
acid of $\frac{1}{6}$th per cent. aqueous solution was then allowed to
act on the Leech for eighteen to twenty-four hours. The
specimen was next removed to half a pint of alcohol (60 per
cent.), and removed to a second half pint on the following
day. After three or four days in the second quantum of
alcohol sections should be cut and stained. The staining
is more successful in recently hardened specimens than in
those which have been preserved many months. Chromic
acid was preferred for the hardening process to Kleinen-
berg's picrin solution or to pure alcohol.

The sections were cut by the aid of a simple screw micro-
tome, and from forty to sixty could be obtained and pre-
served in their order of sequence in the region of a single
pair of nephridia.

The sections were stained in some cases with hæmatoxylin, but usually with Ranvier's picrocarmine, which has a remarkable effect upon the naturally red-coloured hæmal fluid. This very frequently is stained yellow by the picrin alone, whilst the surrounding tissues have a more or less complete carmine staining. Some very beautiful preparations were obtained by Mr. J. E. Blomfield by staining with two pigments in succession, viz. picrocarmine and anilin-blue. Oil of cloves and Canada balsam were used for clarifying and mounting.

Camera lucida drawings, under a moderate power of the microscope (Hartnack's obj. 4), were now made of a *complete series*, giving all the sections in order in which any part of a selected nephridium was involved. Such a series of drawings compared with the whole nephridium, and with horizontal and transverse sections, has enabled me to thoroughly explore this somewhat tortuous and complicated body.

III.—FORM AND REGIONS OF THE NEPHRIDIUM.

The nephridium of the medicinal Leech is not the simple loop-like body with labyrinthine duct opening into a vesicle which it has been held to be, and such as it was drawn by Gratiolet. In the testicular region of the body the nephridium has the form which I have diagrammatically represented in Plate XXIV, fig. 1. It may be divided first of all into *vesicle* and *gland*, which are connected by the *vesicle-duct*.

The gland is in the form of a thick made horse-shoe, the two limbs of which are elongated and produced till they meet. The front of the horse-shoe is dorsal and superior in the living Leech (to the right in the figure), whilst the produced limbs descend and are respectively anterior and posterior in position; in the ventral region they are twisted out of the straight line in a forward direction (see figure). From the forwardly placed thickened point of union of the two limbs of the horse-shoe reaches a delicate process which I call the TESTIS LOBE. It is more or less rudimentary in those nephridia which belong to the anterior region of the body where there are no testes, the further forward the nephridium the smaller is the *testis lobe*.

The vesicle relatively to the form and position of the horse-shoe with its prolonged and bent limbs may be said to *balance* the testis lobe. It lies posteriorly and ventrally rela-

tively to the main curve of the horse-shoe in the living
Leech.

The vesicle duct, instead of passing into that limb of the
gland which is the nearer to it, crosses the posterior limb
externally, and joins the anterior limb. A portion of the
gland, undescribed by previous observers, is found in the
form of a delicate cord of gland-substance which extends as
a free piece (a sort of third limb) from the centre of the
concavity of the horse-shoe to about the middle of the
posterior limb to which it is united.   This is marked in the
figure by the words " Recurrent Duct."

It would not be difficult to give names to the different
parts of the elongated horse-shoe shaped mass just described
which might serve in further description did the minute
structure of the gland in any way correspond with the super-
ficial appearance.   As a matter of fact it does not, and I
have found it necessary to separate the following regions or
lobes which have little correspondence with the general
external form.

The whole curve of the horse-shoe, from the point where
the vesicle duct enters the anterior limb to the point where
the recurrent duct joins the posterior limb, may be known as
the MAIN LOBE.

From the point where the recurrent duct joins the posterior
limb to the point where the two prolonged and forwardly-
bent limbs unite, including a portion of the anterior limb,
is the APICAL LOBE.

The outstanding process of the nephridium which comes
into close relation with the testis, and which is given off
from the anterior limb close to the enlarged commencement
of the apical lobe, is the TESTIS LOBE.

The little piece of the anterior limb extending from the
base of the testis lobe to the commencement of the main
lobe I shall leave for the present without a name, whilst the
delicate piece depending from the main lobe and in which
the recurrent duct commences may be called the recurrent
" piece ; " it can hardly be called a lobe.

IV.—THE DUCTS (VESICLE DUCT, CENTRAL DUCT, AND RECURRENT DUCT).

The divisions of the glandular portion of the nephridium,
which we have just recognised, are necessitated by the dis-
position of the duct and by the characters of the cells sur-
rounding it.

The duct in its various divisions appears as a cylindrical
passage filled with a colourless liquid, and often contains

needle-like crystals (fig. 11), which are also found in great abundance in the vesicle.

The VESICLE DUCT is a tube $\frac{1}{400}$ inch in diameter, with thin walls, formed by numerous cells, several cells surrounding the lumen of the tube. It plunges into the substance of the anterior limb of the gland and assumes a different character. Henceforth I call it the CENTRAL DUCT.

The central duct is circular in transverse section, varies from $\frac{1}{200}$ to $\frac{1}{1000}$ inch in diameter, and is lined by a very strongly-marked structureless cuticle, which has a radiating fibrous periphery, the fibrous irregular radii passing into the substance of the nephridial cells which surround the duct, and also passing *between* them, where it becomes the proper cuticle of the cells (see figs. 7 and 13, C. D.). The central duct takes a course in the centre of the mass of cells which constitute the lobes of the gland, and so far from being labyrinthine, is entirely simple and unbranched, excepting at one point where it gives off a lateral offshoot similar to itself.

Previous observers appear not to have distinguished between this central duct, which is the direct continuation of the vesicle duct, and the immense plexus of *ductules*, to be described below, which excavate the cells surrounding the central duct, but which most assuredly are *not* ramifications of the central duct, and which I have not been able *definitely to trace into communication with it at any point*, though I consider it possible that such a communication may exist at some one or possibly two points which have eluded my constant and very careful search.

The central duct at its origin from the vesicle duct turns upwards (to the right in the figure) and traverses the arch of the main lobe, where it is surrounded by the peculiar nephridial gland-cells, which are in parts two or three rows deep. It descends in the posterior limb of the main lobe and passes on into the apical lobe.

At that point of the posterior limb where the apical and main lobes are joined there is a constriction of the whole gland, and a sudden and marked change in the character of the nephridial cells. This sudden change has no effect at all upon the character of the central duct, which pursues its course along the axis of the apical lobe, arriving at last at the recurved apex of that lobe, where it rests upon the root of the testis lobe (see Plate XXIV, fig. 1). At the same time, although the central duct passes across the junction of main and apical lobes without any change, it gives off a branch—similar in character to itself—as soon as it has entered the apical lobe. This branch I term the RECURRENT

DUCT. This recurrent duct takes an axial course in that delicate cord of gland-cells which depends from between the two limbs of the arch of the main lobe, and which, as an inspection of the diagram (fig. 1) will show, is nothing more than a free or detached continuation of the apical lobe beyond the point where it meets the main lobe.   The recurrent duct thus reaches the middle point of the concavity of the main lobe, when it turns downwards again, running among the cells of the anterior limb of the main lobe parallel with the first part of the central duct.   It crosses without any junction the origin of the central duct from the vesicle duct, and runs up the somewhat narrow portion of the gland which connects the main lobe with the base of the testis lobe and apex of the apical lobe.

Here (and I must beg the reader to follow my description with the aid of the figure) the recurrent duct branches, just in the same way as the central duct branched in giving origin to it.   One branch runs up into the testis lobe for a short distance, and there appears to terminate—how I have not been able to determine—but *possibly* this is a point at which the duct and the ductules of the gland are in communication.

The other branch runs into the apical lobe and joins the central duct, as shown in the diagram (fig. 1), and in fig. 2, which is a careful drawing from a fresh preparation of this part.   Thus we find the recurrent duct, which originated as a branch of the central duct at the other end of the apical lobe, coming again into full continuity with the central duct after a long course.

The interpretation of the duct of the Leech's nephridium, with its central and recurrent portions, is exceedingly difficult.   It is totally unlike the duct or central lumen of the nephridium of the earthworm.   Possibly the recurved apex of the apical lobe represents the funnel-like extremity of the nephridium of Nephelis and Oligochæta.   But in the latter the funnel-like aperture leads into a passage which is *not* intercellular as is that of Hirudo, but *intra*-cellular, as are the ductules of the latter.   It is at this apical region—where the recurrent duct rejoins the central duct—that I have looked most carefully for rudiments of the funnel-like aperture, or for a small opening to the body cavity; but I am able to state definitely that none such exists.   The relation, or rather the difficulty of explaining the relation, between the nephridium of the Leech and that of the Oligochæta will become more apparent when we have examined the ductules.

V.—The Ductules and Nephridial Cells.

When I commenced the study of the Leech's nephridium I regarded the numerous minute passages which give the labyrinthine character to that organ noted by Gegenbaur as necessarily only branches and ramifications of the system of the central duct. Nevertheless, in none of the many hundred sections of the nephridium which I have examined have I been able to find a single instance of a ductule opening into the central duct or into its recurrent branch.

The ductules of the Leech's nephridium are passages lined by a firm resistant cuticle, which excavate the secreting cells of that organ in such a way that every individual cell is completely bored through by such a passage (the grey network in fig. 1). Further, the passages differ very much in calibre in the different lobes of the nephridium, and not only that, but differ further from one another in being in some cells simple traversing passages (A. L. in fig. 13), whilst in other cells they are *branched* within the cell (figs. 5, 6, 7). Further still, the branches may be all equally *passages* leading into corresponding passages in neighbouring cells (fig. 3), or most of the branches may be *cæcal*, in which case they are often exceedingly minute (fig. 5).

Nothing equalling in complexity the system of intracellular passages or ductules of the nephridial cells of Hirudo has hitherto been described by previous writers on that animal, nor has anything quite parallel been described in any other cell-structure, so that the facts which I have ascertained with regard to these ductules in the Leech's nephridium have a certain histological interest of a general character.

It was, I believe, first pointed out by Claparède, who for the first time made sections of the nephridium of the earthworm, that the tube of which that organ consists is built up by a single series of cells (throughout the greater part of its length), and that these cells are simply bored through by the duct or lumen of the tube. Each cell accordingly has the form of a drainpipe. A similar structure has now been recognised in the embryonic nephridia of larval Pulmo-Gasteropods (Rabl), and is clearly enough characteristic of nephridia of a certain grade of elaboration.[1] At the same

[1] It is suggested by Professor Lankester that the structure in question probably points to the phyletic origin of the nephridia from simple unicellular glands, rather than from pockets or invaginated pouches of the epidermis consisting of many cells surrounding a cavity. A nephridial tube, consisting of a lineal series of perforated cells, is simply a unicellular

time it is not by any means always exhibited by nephridia, *e. g.* those of adult Mollusca (organs of Bojanus) and those of Vertebrata. I am not aware that a branched duct has been described as excavating the cells of any nephridium, or, indeed, of any gland previously to this. The cells from the nephridium of Hæmopis, figured by Leydig (*loc. cit.*), are simple cylindrical cells with unbranched passages passing through them, and probably come from the walls of the recurrent duct. In the salivary glands of some Insects and Crustaceans the nearest approach to the kind of relation now established between cell and duct in the nephridia is to be found,[1] but I believe that hitherto only an excavation of the gland-cell by the ductules of origin has been observed in these structures, and not an actual thorough perforation of the gland-cell.

I have been able to satisfy myself, by repeated observations, that the whole system of ductules in the nephridium of the Leech is a continuous network of passages, but I have not been able to determine any aperture in that network by which it communicates either with spaces in the general substance of the body or with the central duct of the nephridium, or its branch the recurrent duct. It seems in the highest degree probable that these ductules do communicate with the central duct and so with the vesicle and exterior, yet I have watched them in various conditions of distension, and by pressure have caused certain small corpuscles (fig. 12) which float in the colourless liquid, which more or less distends them, to move along from one ductule to another, without gaining any indication of a communication with the adjacent central duct.

The ductules do not exist in the cells which line the vesicle or its duct. These cells are no doubt homologous with the nephridial cells, yet it is not until the horse-shoe shaped glandular mass is reached that the ductules make their appearance. It is difficult to separate any description of the ductules from that of the nephridial cells themselves, and I shall therefore describe together the appearances presented by these structures in the different regions of the gland.

Cells and ductules of the main lobe (figs. 5, 7, 10, 13).— As in all parts of the gland, the cells are of large size, vary-

gland which has undergone a repeated cell-division transverse to the axis of its duct.

[1] And, it may be noted, that the more archaic examples of salivary glands consist of bundles of unicellular glands, each with its own ductule, which is so far in favour of the hypothesis that the nephridial cells were originally unicellular glands.

ing from $\frac{1}{200}$ of an inch to $\frac{1}{500}$ of an inch in diameter. They
are sub-spherical, tending to the form of polyhedrons, owing
to mutual adpressure. The protoplasm is fairly transparent
in the living state. By the use of reagents it is seen to be
very finely granular, the granular matter being more abun-
dant peripherally, so as to form a cortical layer, which is
strongly marked after maceration in potassium bichromate.
In cells which have undergone prolonged maceration in this
reagent, Professor Lankester observed a rod-like structure
or striation of the cortical layer, the rods being set at right
angles to the lumen of the ductules.

By the use of silver nitrate I have succeeded in demon-
strating a similar structure in the cortical substance of the
nephridial cells from the apical lobe, that is to say, in cells
with a single large traversing ductule (see Pl. XXV,
fig. 9).

A delicate membranous cuticle exists on the surface of
each cell, and is continuous with the cuticle of the ductules
and with the abundant cuticular deposit which forms the
lining of the central duct (figs. 7 and 13, c. D.). The
cuticle of the ductules is well seen in fig. 10. After pro-
longed maceration it is possible to obtain the ductules in an
isolated condition owing to the resistance of their cuticle to
the disintegrating process which affects the surrounding
protoplasm.

Each cell possesses a large well-marked spherical nucleus
varying in diameter from $\frac{1}{2000}$ inch to $\frac{1}{2500}$ inch. The nucleus
has a strongly-marked capsule, and its contents have, after
maceration in weak chromic acid, the appearance shown in
fig. 10 a, viz. a clear substance in which a nucleolar net-
work and nucleolus can be distinguished.

The protoplasm of the nephridial cells of all regions ap-
pears to be distensible and contractile. Whether it is
actively contractile is uncertain, but there is no doubt that
the *volume* of the ductules varies periodically. There is no
muscular coat to the nephridium itself, as there is to the
vesicle, and the change of volume is probably due to the
physiologically changed condition of the secreting cells.
Gegenbaur found the nephridia of Tubifex to be actively
contractile.

The cells are set around the central duct of the main lobe
radially, as many as eight being thus shown in some sec-
tions taken at right angles to the duct. Superficially to
those which thus abut upon the central duct, other cells
similar to the deeper ones may be seen (fig. 13), forming
portions of a second or even of a third row.

In the anterior limb of the main lobe the recurrent duct runs parallel with the central duct, and is in this part of its course surrounded by cells similar to those which surround the central duct adjacent to it.

All the cells of the main lobe have that form of ductule which I distinguished above as *arborescent with some of the branches continuous* (*i. e.* leading into branches of the ductule of a neighbouring cell) *and some of them cæcal.* The best general notion of the arrangement of the ductules and their continuity in the neighbouring cells is given in the upper part of fig. 13. In fig. 10, from a different portion of the main lobe, the arborescent ductules are seen to be limited to one side of the cell, and a tendency to form larger ductules is exhibited. The finest arborescence is that shown in the cell drawn in fig. 5. This is from a particular portion of the main lobe where all the cells have the same character. Five " continuous " branches of the ductule are seen, and a dichotomous series of finer branches which completely honeycomb the cell-protoplasm. The ultimate branches of this system are $\frac{1}{40000}$ inch in diameter. They are so disposed as to open into or (to use another metaphor) to take their origin in the cortical substance of the cell. The rod-like striated structure of this layer has already been mentioned as a general character of the nephridial cells. It appears that the ultimate ramifications of the ductules have a definite relation to the bacillary structure of the cortical substance—although the bacillary structure is observed equally in cells which are provided with a simple in place of a finely arborescent ductule.

The cells of the apical lobe differ from those of the main lobe in possessing very much larger ductules, varying from $\frac{1}{1000}$ inch in diameter to as much as $\frac{1}{500}$ inch, whilst the cells themselves vary from $\frac{1}{200}$ inch to $\frac{1}{250}$ inch in diameter (figs. 3 and 6, and lower part of fig. 13, A. L.). The medium-sized and the largest ductules of this larger sort have a definite position in the gland, the medium-sized gradually passing into the largest as we proceed along the apical lobe *from* the apex towards the posterior limb of the main lobe, where that middle piece with the recurrent duct is given off (see fig. 1). Here the ductules become very wide, and are unbranched, so that the cells are mere hollow cylinders. A good view of the relation of wide-branched ductules to the cells is given in the drawing (fig. 3), which represents cells of the apical lobe in a state of distension. The ductules of the apical lobe are also well seen in fig. 2, A. L.

The ductules of this wider and larger kind are lined with

a very distinct cuticle, which in sections of hardened preparations is often seen in a state of desquamation (fig. 6).

In these large ductules too, in fresh (and therefore still living) excised nephridia, I have studied the contained liquid. It is perfectly colourless and transparent, but frequently contains structureless globules (fig. 12) which, by pressure, can be made to move along the series of ductules.

The cells of the testis lobe (fig. 2, T. L.) agree altogether in structure with those of the apical lobe, having medium-sized ductules which are branched to a small extent, the branches being continuous, not cæcal. In each cell the ductule gives off some two, three, or four branches of the same calibre, and each of these is continuous with a similar branch of the ductule in a continuous cell.

Communications of the ductules of different lobes with one another.—The diagram (fig. 1) will serve most satisfactorily to explain what I have ascertained on this point. The ductules of the testis lobe do not directly communicate with those of the adjacent apical lobe, but run on through the short bit of gland joining testis lobe and main lobe, and then gradually pass into the finer kind of ductule which is found throughout the main lobe. There is no junction at the apex of the main lobe (near the word "inferior vein" in the diagram) between the ductules of the main lobe and the ductules of the apical lobe, which is all the more remarkable since the central duct is here continued from the one lobe to the other. On the other hand, at the point where the "recurrent piece" joins the concavity of the main lobe, as seen in the figure, there is an extensive communication of the large ductules of the "recurrent piece" with the ductules of the main lobe.

VI.—Blood-vessels and Tunic of the Gland.

The mass of cells which forms the glandular portion of the nephridium, with its various lobes, is held together by an investment of fibrous tissue, and is further beset by a large number of blood-vessels which, running longitudinally on the superfices of the gland, give off branches which penetrate the mass of cells at numerous points, and form within the cell-mass *one of the most complete inter-cellular blood-plexuses* known to exist, and resembling that of the Mammalian liver.

The investing fibrous tissue of the gland I shall not describe in detail. It is dealt with by Professor Lankester in an article "On the Connective Tissues of the Leech," published in the present number of this journal. It will be

sufficient to point out that the investing brown-coloured net-
work of fibres consists of what Professor Lankester has
termed " vaso-fibrous tissue," which is found throughout the
body of the Leech, more or less pigmented, and forms the
pigment fibrils which interpenetrate the superficial epi-
thelium of the body-surface.

In the transverse section (fig. 13) this tissue is seen as
dark granular matter (f) surrounding the lobes of the gland,
and passing in and out amongst the muscular fibres (g) and
the unicellular glands (e), all of which are more or less
closely invested by it.

The peculiarly close relationship of this tissue to the
capillary blood-vessels themselves explains how it is that no
distinct connective tissue penetrates the cell-mass of the
nephridium *but only fine blood-vessels*, which form, in fact,
an intercellular connective tissue, which is at the same time
a capillary system.

The large blood-vessels found in connection with the
nephridium have been described with great care and ac-
curacy by Gratiolet. They are as shown in fig. 1. They
consist of vessels with delicate membranous walls which are
given off by the great lateral muscular vessel passing near
the nephridium. A connection is further established between
the capillary plexus of the nephridium and the dorsal longi-
tudinal vessel (which runs along the dorsal surface of the
alimentary canal) by means of the "superior vein." The
arrangement and distribution of the vessels will be best
understood from an examination of the diagram fig. 1. The
smaller vessels which are arranged so as to form a plexus or
meshwork with a single nephridial cell in each hole of the
meshwork average in size $\frac{1}{1000}$ of an inch, but are capable
of being greatly distended with blood, and do not always or
in all parts of the nephridium appear to be fully developed.

This, however, is not due, I believe, to any structural
irregularity, but to a functional state. When the nephri-
dial cells or their ducts are swollen, the intercellular vascular
network is compressed, and many of its channels are tem-
porarily obliterated. But, on the other hand, it may happen
that we find all the blood-capillaries well injected with
their natural red-coloured fluid, as shown in figs. 6 and 7.
These are drawn from such " natural injections " which ex-
hibit themselves in the most beautiful way when sections
are cut from specimens of the Leech taken in full nutrition
(that is, within a week or so of a good meal), and hardened
in dilute chromic acid.

The wall of the finer blood-vessels running between the

nephridial cells is perfectly distinct. These are *not* mere intercellular passages but have their distinct membranous walls like those of the finer blood-vessels in other parts of the Leech. In these thin-walled vessels, both in the nephridium and elsewhere, I have not succeeded in tracing any cell-structure by the use of various histological methods. It appears propable from what Professor Lankester has ascertained with regard to the relations of the vaso-fibrous tissue and the blood-vessels, that the nuclei are discharged at an early period of its development from the wall of the blood-vessel into the lumen of the vessel, and accordingly we find only the metamorphosed cell-substance left to form the wall which is accordingly structureless.

VII.—The Vesicle and the Secretion of the Gland.

The vesicle is remarkable for its great dilatability, and for possessing a muscular coat which the glandular portion of the nephridium does *not* possess. On this latter point I am at variance with Professor Leuckart, who has ascribed a muscular coat to the glandular portion. The vesicle may be regarded as an expansion of the central duct to which it is joined by the " vesicle duct," but it differs greatly from any part of the glandular portion of the nephridium, not only in the fact that it has a muscular tunic, but also in the fact that the cells lining it have *not a cuticle* but a *ciliated surface*. Cilia are not found in any other part of the nephridium, which is a remarkable fact, when we compare the structure of other nephridia such as those of the earthworm and the molluscan organ of Bojanus. The cilia on the epithelium of the vesicle are exceedingly short, the cells themselves being small and short, and thrown into ridges when the vesicle is in a state of contraction. The vesicle is seen in section in the drawing (fig. 14), which represents accurately an actual preparation.

The vesicle does not open directly to the exterior, but is placed in communication with the body-surface by a short duct, which is lined near its external opening by an involution of the epidermis of the integument.

The wall of the vesicle is supplied with a very regular capillary plexus of the hæmal system (fig. 1), and it seems probable that secretion may be carried on from its walls as well as from the more distinctly glandular portion of the nephridium.

The vesicle contains a liquid in which bunches of needle-like crystals are found, which are drawn in fig. 11, as seen when magnified 3500 diameters by means of Hartnack's

objective " 10 immersion." These crystals are not stained by iodine ; they are soluble in nitric acid. I have not yet completed their chemical examination. These crystals are occasionally to be found in the central duct, but I have never found them in the ductules. On the other hand, in the ductules are found minute, structureless corpuscles (fig. 12), which on one occasion I saw exuded from the substance of a nephridial cell into the ductule. I have never found these corpuscles in the central duct nor in the vesicle.

VIII.—Opening of the Systems of Ductules and Ducts into one another and into the Body-Cavity.

As I have before mentioned, I have failed to establish the existence of a communication between the central duct or its off-sets and the system of ductules ; but I am strongly inclined to regard the recurrent duct as the seat of this communication. It seems to me not improbable, from the large size of the ductules of this portion, and from the way in which it enters the apical lobe on the one hand and the concavity of the main lobe on the other, that we shall eventually find a connection through it of the two systems.

I hope during the present summer, by the study of other genera of Leeches (Aulostomum, Nephelis, Clepsine, Pontobdella), to gain some light upon this question.

The absence of cilia from the ducts and ductules of the Leech's nephridium, together with the absence of any large space in the body—comparable to a body-cavity—seems to be correlated with the absence of an open internal termination to the duct and ductule systems. I am inclined to regard what I call the apical lobe as probably corresponding with the position in which an internal opening is found in the typical form of nephridium, such as occurs in Nephelis, Clepsine, and the Oligochæta.

This point also will be, I hope, decided by the investigations which I am now commencing on those genera.

The very wide difference between Hirudo and Clepsine (so far as descriptions of the latter genus enable me to judge) in the whole arrangement of duct and nephridial cells leads me to believe that the comparison of the series of Hirudinean genera will be a very interesting one.

SUMMARY.

1. The Leech's nephridium consists of gland and vesicle connected by the vesicle duct.

2. The gland is a curved, horse-shoe shaped loop, present-

ing the following *lobes* :—1, main lobe; 2, apical lobe; 3, testis lobe; 4, recurrent lobe.

3. The cells which constitute the gland are all penetrated by *ductules,* which pass through them, sometimes forming several passages; the ductules differ in the different lobes of the gland.

4. The axis of all the lobes is occupied by a large duct separated from the nephridial cells by a thick cuticle; it opens into the vesicle duct, but is not shown to have any connection with the ductules.

5. Vessels of the hæmal system (with red fluid) form a complete plexus in the gland, each cell being surrounded by a loop of the plexus.

6. There is no internal opening of the system of ducts or ductules.

7. There are no cilia in any part of the gland excepting the vesicle, which is lined by a ciliated epithelium.

8. The wall of the vesicle is muscular; that of the gland is not, but consists of a fibrous, pigmented investment.

On Intra-Epithelial Capillaries *in the* Integument
of the Medicinal Leech. By E. Ray Lankester,
M.A., F.R.S., Professor of Zoology in University Col-
lege, London.   With Plate XXVI.

I am not acquainted with any minute investigation of the
structure of the epidermis of the common Leech.   Professor
Leydig has published most valuable accounts of the sense-
organs of the Leeches, and in a drawing of a transverse sec-
tion of the medicinal Leech (' Bau des Thierischen Korpers,'
Taf. i, fig. 6), has shown the epidermis in position with the
remarkably enlarged cells which form unicellular glands
plunging down amongst the muscular bundles far below the
horizon of the other epidermal elements.   The same draw-
ing also shows a few scattered vessels belonging to the red
vascular system.   These are seen in Professor Leydig's draw-
ing to come near the integument, but they do not penetrate
it.   The drawing is on a relatively small scale, so that such
details as the minute structure of the epidermic cells are not
presented.

In sections prepared in my laboratory last year by Mr.
Blomfield and by Mr. Bourne, and in macerated specimens,
I have studied the structure of the Leech's epidermis more
closely, and have obtained some interesting results.   I was
led to make these observations in carrying out a general plan
of study of the epidermic tissue in the various groups of the
animal kingdom.   The integument of the Earthworm two
years ago yielded me some very interesting results entirely
contradictory to Claparède's statements on that subject—more
delicate and varied forms of epithelial cells than are yielded
by the Earthworm's epiderm when macerated are not pre-
sented by any other animal—whilst the whole structure of
the clitellum and its periodical variation is especially in-
teresting.   Amongst the most important facts established
in regard to the Earthworm's epiderm are, firstly, the existence
beneath the cuticle of a normal cellular matrix, consisting of
varied forms of goblet cells and excessively delicate elongate
interstitial or " packing " cells, instead of the altogether im-
probable syncytium described by Claparède ; secondly, the
penetration in the region of the clitellum of vessels of the
hæmal system in the form of loops between the groups of
epidermic cells ; thirdly, the similar penetration of processes
of pigment cells belonging to the connective-tissue system
among the epidermic cells of the general body-surface (espe-

cially in *Lumbricus olidus*). The publication of these results was delayed by other work, which is the less to be regretted since Dr. Horst[1] (1876) and Dr. Von Mojsisovics [2] (1877) had but a short time previously applied themselves to the same investigation and arrived at results of a similar character, though their memoirs had not come into my hands until after I had completed my examination of the Earthworm's integument. The drawings of Dr. Von Mojsisovics do more justice to the very beautiful structure of the Earthworm's epiderm than those of Dr. Horst, at the same time there are still some points not fully illustrated or appreciated by the former which I hope to see treated in this journal by one of my pupils.

The epidermis of the Leech contrasts very strikingly with that of the Earthworm.

The cells which constitute it are very nearly uniform in size and columnar in shape ($\frac{1}{1500}$ inch long $\frac{1}{10,000}$ inch broad on the average).

Gland Cells.—Only a few epidermic cells are enlarged to form glands, and these do not as a rule remain in the horizon of the other cells but sink below the surface, having only a narrow duct to represent them in the epidermic stratum.

These unicellular glands may be roughly divided into two series, those which are more superficial and those which occupy a very deep position. Some of the very deep-lying epidermal gland cells are seen in Mr. Bourne's drawing of a section of a nephridium (Pl. XXV, fig. 13 *e*), whilst a more superficial one is seen in (Pl. XXVI, fig. 3 *gl*). I have not made a special study of these gland cells, which would well repay the attention of the physiologist, especially that variety of them which opens into the stomodæal invagination of the epiderm, and which is known as salivary gland. The small and more superficial gland cells appear to be most abundant about the generative region of the body, where they may occur so thickly as one to every six or seven columnar cells when seen in transverse section. There is little doubt that the superficial series of gland cells serve, in this region, to secrete the material of which the egg-cocoon is formed.

Cuticle.—External to the epithelium of the body wall is a continuous cuticle, which is more delicate than the corresponding cuticle of the Earthworm, and quite devoid of striations or lamination. It exhibits at intervals (fig. 5)

[1] 'Aantcekeningen op de Anatomie von Lumbricus terrestris,' Utrecht, 1876.

[2] " Die Lumbriciden hypodermis," 'Sitzber. Akad.,' Wien, 1877.

perforations which are irregularly placed but tend to a disposition such as that seen in the figure.   These perforations are the openings of the unicellular glands.   Frequently two such apertures and sometimes three are placed closely side by side.

Columnar Cells.—When the epithelium is carefully examined by means of very thin sections and by teazing, it is found that the columnar cells which form the stratum subjacent to the cuticle have not a simple columnar form but are T-shaped or rather mallet-shaped (fig. 1).   The broad expanded portion, the " head " of the mallet, forms with its fellows a complete mosaic (fig. 6) immediately underlying the cuticle, whilst the "handles " of the mallets are arranged side by side so as to leave a certain amount of space between neighbouring "handles" (fig. 3).   Into these spaces processes of the connective tissue here and there make their way, these processes being outgrowths of the pigmented vaso-fibrous tissue which pervades the whole body of the Leech (fig. 3 *pg*).   The variously coloured spots of the Leech's integument are produced by modifications of this tissue, and the amount of it which interlocks with the epithelial cells varies from point to point according to the pattern of the Leech's colouring.

Columnar cells which have been teased out, after maceration of the Leech in potassium bichromate, show the expanded " head " having a finely granular or sometimes vacuolated structure, whilst the "handle " appears very nearly homogeneous (fig. 1).   I have not been able by staining agents to show a nucleus in any part of the cell.   The whole of the " handle " takes the staining rather freely, and it is possible that it consists chiefly or entirely of nucleus.

When a detached flake of epithelium, which has been stained by picro-carmine, is looked at from above, the appearances represented in figs. 6, 7, 8, 9 are obtained.   A complete mosaic of polygonal cells is seen, in *some* of which are dark, that is to say, well-stained bodies, which look like nuclei.   In others of the polygonal area there are no such darkly-stained bodies, but only fainter indications of a differentiation.   The result is that the cells with darkly-stained bodies in them are arranged in irregular groups (fig. 6).   The darkly-stained bodies prove upon further examination to be *not* ordinary nuclei, but the " handles " or depending portions of the epithelial cells.

In such flake-like preparations some of the cells are seen to be perforated by one or by two circular holes.   These are the passages for the ducts of the unicellular glands.

A similar perforation of superficial epidermic cells for the transit of ducts of subjacent glands is seen in the skin of the common frog.

Intra-epithelial Blood-capillaries.— In sections from any region of the Leech's body the fine vessels of the hæmal system naturally injected with their own hæmoglobin-coloured fluid may be traced up to the sub-epidermal connective tissue, and thence finer branches may be here and there followed into the spaces between the " handles " of the mallet-like epithelial cells. Three examples of this disposition of structures are given in Plate XXVI. Fig. 2 is drawn from a very thin section, which allows the observer to trace a horizontal vessel threading its way among the columns of the epithelium. In fig. 3 a section is shown which has fortunately cut one of the intra-epithelial vessels transversely, whilst in fig. 4 a number of vessels are seen supplying the intra-epithelial plexus, the section being too thick to show well the individual form of the epithelial cells.

The sections shown in these three figures are merely samples of a perfectly general arrangement which occurs over the whole surface of the Leech's body. The intra-epithelial vessels form a continuous plexus, giving to the whole of the epithelial layer of the skin a vascular character.

General Remarks.—As a histological fact, the vascularity of this epithelium is a novelty. The only vascular epithelium, so far as I know, which has been hitherto described, is that of the clitellum of the Earthworm ; but in that case the vessels do not penetrate between the cells of the most superficial layer.

The penetration into the epithelium of the processes of the pigmented vaso-fibrous tissue is a phenomenon which might be expected to accompany the penetration of the vessels, since the vaso-fibrous tissue and the capillary blood-vessels are virtually one and the same tissue.

Physiologically the significance of the arrangement I have described is obvious enough. The true respiratory organ of the Leech is clearly this vascular epiderm, and amongst respiratory organs it stands alone in the nearness with which the absorbent blood-vessels succeed in bringing themselves through all structural obstacles into direct contact with the oxygenating medium.

On the CONNECTIVE *and* VASIFACTIVE TISSUES *of the* ME-
DICINAL LEECH.[1]   By E. RAY LANKESTER, M.A., F.R.S.,
Professor of Zoology and Comparative Anatomy in
University College, London.   With Plates **XXVII** and
XXVIII.

THE minute comparative anatomy of that group of tissues
which is more or less vaguely indicated by the term " con-
nective substance " (with which I should associate—as is not
usually and expressly done—the vasifactive [angioplastic]
derivatives of connective tissue[2]) has yet to be worked out,
and when the task has been accomplished the great " con-
nective-tissue question," in which histologists were but lately
so keenly interested, will be settled.

There can be no doubt that from the point of view of
general morphology, as well as from the more special point
of view of the histologist, the proper understanding of the
nature and relations of the varieties of connective and vasi-
factive tissue is of fundamental importance.   You have in
the ideal connective substance an element the vast possi-
bilities of which first impressed the mind of Bichat—an
element which can assume the widest variety of form and
function, embracing the conditions known as tendon, fat,
capillaries, cartilage, areolar tissue, and endothelium.

To study the origins of this tissue and its varieties in
lower forms of organisation cannot but be a fruitful labour,
since the histological differentiation of the higher Vertebrates,
no less than their grosser morphological composition, must
be traced, if we are rightly to comprehend it, to simpler and
more archaic phases which are to some extent at least pre-
served in those branches of the great animal pedigree which
have not advanced so far from a primitive ancestral condition
as have the Vertebrata.

Ectoplastic and entoplastic tissues.—The most fundamental
distinction which can be drawn morphologically between the
various kinds of connective tissues, is one which also serves
to divide other groups of tissues and depends on the mode

[1] See also ' Zoologischer Anzeiger,' 1880, No. 49.
[2] The term " connective tissue " is used very unfortunately in differing
degrees of comprehensiveness—sometimes cartilage, sometimes fat, and
sometimes bone, being excluded or included as the case may be.   I would
propose the term " skeleto-trophic " for a natural group of tissues which is
divisible into—(1) Skeletal, including fibrous, adenoid, adipose, bony, and
cartilaginous tissues. (2) Vasifactive, including capillaries and embryonic
blood-vessels.   (3) Hæmolymph, including the hæma or hæmaglobinous
element and lymph, the colourless element of vascular fluids.

in which the metamorphosis of the embryonic protoplasmic cells, from which all tissues are developed, commence that differentiation which leads to the permanent form of the tissue which they constitute. The mother-cells of all tissues are either "entoplastic" or "ectoplastic," or both—that is to say, the metamorphosis of their protoplasm is either essentially one occurring at the surface of the protoplasmic corpuscle, or one occurring deeply within its substance, or the two processes may go on in connection with the same cell.[1] As examples of the two classes I may cite fat-cells as essentially entoplastic, and the corpuscles of areolar tissue as essentially ectoplastic. No doubt both intra-cellular and inter-cellular deposits do occur in one and the same tissue, but in most tissues it is possible to point to one or other mode of deposition, or of metamorphosis, as that which is characteristic. Tissues which present both phenomena in a nearly equal proportion may be distinguished as "endecto-plastic." In the group of the connective tissues we find that the very same chemical and physiological result may be obtained by a metamorphosis which is either entoplastic or, on the other hand, ectoplastic. Thus hyaline cartilage is essentially ectoplastic, whether we take the well-known type with rounded corpuscles embedded in a matrix, or the more unusual form with arborescent cells found in the cartilages of fish and of Cephalopoda. On the other hand, notochordal tissue, which is often loosely spoken of as " a kind of cartilage," differs profoundly from ectoplastic cartilage in the fact that the cell metamorphosis is essentially entoplastic, occuring *within* the area of the original protoplasmic corpuscle, and not in the form of a deposit surrounding and embedding the embryonic unit. In that variety of connective tissue which chemically differs from cartilage in yielding a less dense and resistant product of metamorphosis than that which is associated with the name of cartilage, we find equally the two great morphological varieties of ento-plastic and ectoplastic tissue. Fibrous tissue generally is ectoplastic—that is to say, the protoplasmic corpuscles remain more or less intact whilst surrounded by the fibrous and lamellar masses to which they have peripherally or laterally given origin. This is true of ordinary subcutaneous areolar tissue, of tendon, of mucous tissue (umbilical cord, &c.), and of corneal tissue. At the same time we find in various Invertebrate groups (not in the Vertebrata) an

[1] When this is the case the ectoplastic and the entoplastic products of metamorphosis are usually of a different chemical nature and of different physical properties.

entoplastic form corresponding chemically and functionally to the ectoplastic forms just cited. This is the " grossblasige Bindegewebe "—the vesicular connective tissue so abundant in the Mollusca, in the Nemertines, and other Invertebrata. The only tissue which in *form* represents this among the connective tissues of Vertebrata is adipose tissue. Vesicular connective tissue stands in the same relation to mucous tissue, tendon, and corneal tissue, as does notochordal tissue to hyaline or fibrous cartilage.

Yet further, the tissues of the connective group which are specially related to the nutrient fluids (such as blood and lymph), and which form the wall of the cœlom or of blood-channels, may be entoplastic when they give rise by internal metamorphosis (liquid vacuolation) to capillary vessels, or ectoplastic when they constitute spongy or lacuniferous cell aggregates, the cells separated by *inter*-cellular channels, such as we find in the " pulp " of lymph-glands and the spleen and in the lacunar tissue of Molluscs.[1] The recognition of these two modes of cell metamorphosis appears to me to be important, as enabling us to bring together as morphological varieties of one and the same tissue, having the same chemical and physiological significance tissues, which are frequently separated from one another without scientific method, or else are abitrarily placed side by side in consequence of the recognition of chemical and physiological resemblances, without sufficient emphasis being given to their very different morphological character.

Ectoplastic and entoplastic connective substance of the Leech.—In the medicinal Leech two forms, and only two chief forms of skeleto-trophic tissue are to be discovered. The one is an ectoplastic connective substance resembling somewhat the jelly-like tissue of the umbilical cord (Pl. XXVII, fig. 1); this I shall term " ectoplastic connective jelly " (in contrast to the " entoplastic connective jelly " so abundant in Molluscs). The second kind of skeleto-trophic tissue found in the Leech consists of greatly branching and anastomosing fibres (Pl. XXVII, fig. 1, FB, and fig. 3), which are often darkly pigmented by fine granules, and are not unfrequently tubular (fig. 4), passing by insensible gradation and actual continuity of substance into the form of very delicate capillaries, which abound in all parts of the Leech's body and contain hæma, that is, fluid impregnated with hæmoglobin. It is a modification of this tissue which forms the pigment patches of the integument (see Pl.

[1] See R. H. Peck, "On the Structure of the Lamellibranch Gill," this Journal, 1876.

XXVI, fig. 4), and an extreme modification in the opposite direction, which appears under the form of botryoidal tissue (Pl. XXVIII, fig. 15) called hepatic by Brandt, mistaken for cutaneous glands by Leuckart, and compared to the fatty body of insects by Leydig. I propose to term this ento-plastic form of skeleto-trophic tissue—which has so Protean a character—"vaso-fibrous tissue."

The connective jelly of the Leech.—This tissue may be best studied on perfectly fresh samples, which are to be obtained by teasing any portion of the muscular paren-chyma which intervenes between the integument and the wall of the alimentary canal.

It consists of a jelly-like matrix, in which are embedded nu-merous branched corpuscles, varying in diameter from $\frac{1}{4000}$th to the $\frac{1}{3000}$th of an inch.[1] The branches of the corpuscles are drawn out into very fine processes, and finally are lost in exceedingly delicate filaments, which permeate the jelly in every direction. Treatment with nitrate of silver gives the usual result observed with such tissues, viz. a dark staining of the matrix, whilst the corpuscles are left as clear spaces.

The corpuscles vary in size and in colour in different specimens of the tissue. When young they are small and contain a few highly refringent granules of large and regu-lar size. Older tracts of the tissue show corpuscles of larger size, the granules within them being very numerous, of large size, and similar to one another; the granules are in these older cells of a decided brown or straw colour. In some parts the corpuscles may be found to be very much elongated, and suggests the possibility of a transition from the ecto-plastic connective jelly to the entoplastic vaso-fibrous tissue.

Even in perfectly fresh corpuscles of the connective jelly a clear space amongst the granules may be observed, which indicates the nucleus. Treatment with osmic acid, followed by picro-carmine (on the glass-slip, whilst under a cover-glass, in the manner recommended by Ranvier), gives two interesting results. First, the granules become very much stained by the osmic acid, and, secondly, the nucleus, but no other part of the corpuscle, becomes coloured by the carmine (Pl. XXVII, fig. 2).

The highly refringent granules of these corpuscles are very characteristic on account of their largeness, their colour, and their regularity of form and size. They are not simply fatty matter, since they are not dissolved by ether. They are possibly of the same chemical nature as the very much

[1] A very similar tissue occurs beneath the epidermis of *Sipunculus nudus,* having, however, a more cartilaginoid matrix.

more minute brown-coloured granules which occur in various parts of the vaso-fibrous tissue.

The vaso-fibrous tissues of the Medicinal Leech.— Whenever any portion of the muscular tissue or any organ of the leech is examined under the microscope, a large quantity of dark brown fibres are sure to be noticed, either forming an investment to the organ or ramifying over the field of the microscope (Pl. XXVII, fig. 3).

This system of fibres penetrates every region of the Leech's body. It is continued superficially into the epidermis, beneath and between the cells of which it forms the characteristic pigment spots. It occurs between the bundles of muscular fibres, as may be seen in the section (fig. 13 *g.*) of Mr. Bourne's Pl. XXV. It forms a loose sort of feltwork on the surface of the nephridium of the testes and of the alimentary canal. It is particularly rich and thickly developed on the wall of the blood-sinus in which the ventral nerve-cord is placed.

When examined with a high power the fibres are seen to be of varying thickness, and many of them apparently hollow, so that the larger fibres are tubular rather than solid, and moreover, the tubular wall, according to the condition of tension or relaxation of the tissue, may be smooth or thrown into transverse rugæ (Pl. XXVII, fig. 1). Treatment with osmic acid, followed by picro-carmine, has the effect of demonstrating the nuclei of the cells by which this tissue is formed. The nuclei (Pl. XXVII, fig. 4) are seen to be scattered at intervals in the course of the fibre, and when this is tubular they project internally into the lumen of the tube. The substance surrounding the nuclei is densely packed with very fine granules and does not stain red with picro-carmine as do the nuclei themselves. In many fibres (apparently old and fully differentiated) nuclei are either exceedingly rare or are not to be detected. It appears probable that in these fibres the nuclei of the original cells have disappeared and left only the finely granular cell-substance to play the part of a connective substance.

In many of the tubular fibres, however, the nuclei are abundant, and show a series of transitional conditions, indicating that they gradually become freed from the granular cell-substance, and at first project into the cavity of the tube, and then become detached and float freely in it (Pl. XXVII, fig. 4 and fig. 5). Some of the fibres of this system, which are definitely continuous with the darker and thick-walled fibres, have a pale aspect and relatively few granulations in the wall, whilst the nuclei are abundant

(fig. 5). The appearance of these pale hollow fibres at once impressed me with a close similarity to the developing capillaries of Vertebrata. I had previously been able to demonstrate (see this Journal, vol. xviii, " The Vascular Fluid of the Earthworm a Corpusculated Fluid ") that the delicate vessels of the red vascular system of the Earthworm contain corpuscles, which are nothing more nor less than the liberated nuclei of cells which line the walls of those vessels, and, accordingly, I was prepared to find in the vessels of the Leech (in the congeners of which the existence of " blood-corpuscles " has been generally asserted) corpuscles developing by the liberation of nuclei belonging to the cells which constitute the vascular wall.

At the same time the structures which I had before me (Pl. XXVII, figs. 4, 5, 6, 7) were not vessels forming part of the red vascular system of the Leech, but tubular fibres connected with the general system of brown-coloured fibres pervading the body of the Leech. The vessels, in fact, in which I observed this displacement of the nuclei were not filled with hæmoglobinous fluid, but had clear contents, and evidently had not yet—if ever they were destined to have—any communication with the red-coloured vascular system. My next care, therefore, was to search for any evidence of the connection of such tubular portions of the brown-coloured system of fibres with the blood-vessels.

Thin-walled blood-vessels of the Leech.—The finer vessels of the Leech's hæmoglobinous vascular system are remarkable for the exceeding delicacy of their walls. In no instance have I succeeded, by means of reagents, in demonstrating any nuclei in those walls; and, further, by the use of nitrate of silver I have not succeeded in obtaining any indication of cell-outlines on those walls which I expected to be able to demonstrate since Mr. D'Arcy Power had, when working with me at the histology of the Earthworm, obtained such cell-outlines in the finer vessels of that animal (this Journal, 1878), where, moreover, nuclei are always readily demonstrated, even in the very finest capillaries. The structureless character of the thin-walled blood-vessels of the medicinal Leech is, then, a peculiarity which separates them from the apparently similar vessels of the Earthworm, and renders it not improbable that the two sets of vessels may have a diverse developmental history. Rarely I have found corpuscles consisting of free nuclei in the vessels containing red fluid in the Leech. Two such instances are drawn in Pl. XXVII, figs. 13 and 14. In the one case the

red fluid is still present, in the other the action of reagents had caused its removal, leaving only the corpuscles.

Connection between the brown fibres and the thin-walled vessels.—Here and there a careful search in longitudinal sections of the Leech (hardened in chromic acid, $\frac{1}{4}$ per cent., followed by alcohol) revealed some thin-walled vessels (filled with their red fluid), in which the usual structureless character of the wall was modified. Instead of being absolutely structureless the membranous wall showed thickenings, which were densely packed with fine brown granules, identical with those of the brown fibrous tissue. After some search I found several examples of *the direct continuity of thin-walled vessels with the tubular fibres of the brown-pigmented system of fibres.* Three such instances are represented in Pl. XXVII, figs. 8, 10, and 11. Figs. 9 and 12 represent cæcal terminations of branches of the red vascular system, in which it appears that the cæcal extremity of the branch is continued into a fine fibre. Such an appearance admits of the interpretation that a præ-formed tube has been placed in communication with the red vascular system, which has then filled it with hæmoglobinous fluid up to the point where its lumen ceased, and where the tube ceases to be tubular and is merely a solid fibre.

From these observations I cannot doubt that the thin-walled blood-vessels and the brown-pigmented fibres of the medicinal Leech are virtually one and the same tissue, passing into one another by insensible gradation, and, what is of more importance, actually in continuity with one another at certain points, as processes of one and the same set of vaso-fibrous cords.

It is not possible at present to offer a definite opinion as to whether the development of tubular portions of the brown-pigmented system of fibres into thin-walled vessels is a process which is always and normally going on in the Leech's body. It is possible to suppose that the solid brown fibres, the tubular brown fibres, and the thin-walled hæmatophorous vessels, are three *permanent* varieties of the vaso-fibrous tissue. But I am inclined to think from the fact that not even young hæmatophorous vessels with nuclei in their walls are to be met with in the Leech, and from the fact that the tubular brown fibres discharge into their tubular cavity numerous nuclei identical in appearance with the corpuscles which are here and there to be detected in the hæmatophorous vessels, that there is actually a continual development of offshoots of the brown-pigmented system of fibres into thin-walled hæmatophorous vessels, the nuclei enter-

ing the blood-stream when communication is once esta-
blished and the brown granular wall becoming transparent
and structureless by the absorption of its granules.

Botryoidal tissue (so-called hepatic tissue).—I have now
to notice another development of the vaso-fibrous tissue, the
characters of which are such as to confirm the view already
advanced as to the potential identity of the brown-pigmented
fibres and the vessels of the red vascular system. Nearly
surrounding the wall of the alimentary canal, but separated
from the epithelium of that organ by a vascular plexus, by
a peculiar series of muscular fibres, and by fibres of the vaso-
fibrous system, is a mass of dark-brown botryoidal tissue
(Pl. XXVIII, fig. 15), which has been, on account of its
position and colour, regarded as "hepatic." I was not a
little surprised on studying the structure of this tissue by
means of transverse sections and of teased preparations, to
find that it has in no way the least structural resemblance to
a hepatic gland, but is simply a plexus of hæmatophorous
vessels, the walls of which are swollen in botryoidal fashion by
the enlargement of the individual cells which constitute them.
The cells which constitute the wall are a single series; there
is no external tunic and no internal endothelium. In those
parts of the plexus where its special character is fully de-
veloped every constituent cell is seen to have a hemispherical
shape, standing out on the surface of the vessel (Pl.
XXVIII, fig. 19), and its substance is found to be densely
charged with very fine brown-coloured granules. A clear
nucleus can be detected in most of the cells. The lumen of
the vessels bounded by these singular cells is larger than
that of the average thin-walled hæmatophorous vessel, mea-
suring $\frac{1}{1500}$th of an inch on an average. The vessels form a
true network in the denser part of the plexus, but every-
where cæcal branches of the vessels are found, which appear
to be buds, as it were, of the system, which are growing for-
ward to form unions with other branches, and so extend the
plexus; near the boundary of the masses of botryoidal tissue
very interesting preparations may be obtained, showing the
connection between the botryoidal vessels with thickened
cells to their walls and the thin-walled vessels of the general
hæmatophorous system. Such a preparation is drawn in
Pl. XXVIII, fig. 18. The botryoidal vessels are seen as
buds or branches carried by a thin-walled vessel.

The nature of the granules contained in the cells of the
botryoidal vessels I have not in any way determined, and I
am altogether unable to suggest at present what may be the
significance of this portion of the vaso-fibrous system. It

appears to me that the granules are similar to those of the brown-pigmented fibres, but of this I am by no means certain.

Further, it appears exceedingly probable that these granules are of the same physiological nature as the yellow-brown granules which load the endothelial cells of the cœlom of the Earthworm and other Chætopoda in the region of the alimentary canal. In the Earthworm these cells not only form a complete tunic to the intestine, but also invest the large blood-vessels. At the same time it must be distinctly pointed out that the cells which thus clothe the large vessels of the Earthworm and the contractile vascular cæca of Lumbriculus are *not* the cells which form the proper wall of the blood-vessel, but lie externally to these. Hence their rela · tion to the vascular space and to the hæmoglobinous fluid is essentially different from that of the granular cells of the botryoidal vessels of the Leech.

Previous observations relative to the brown fibres and botryoidal vessels of the Medicinal Leech.—It is not desirable to go over the history of a subject which has not of late years formed the subject of serious investigation. The opinion of Brandt that the botryoidal form of the vaso-fibrous tissue of the Leech was hepatic in character has never been maintained by any histologist. The two observers who have studied the Leech with the aid of the microscope, namely, Leuckart and Leydig, have both rejected Brandt's view as to the botryoidal tissue, but have advanced divergent opinions of their own.

Leuckart (' Die Menschlichen Parasiten,' vol. i) studied the anatomy of the Leech by means of transverse sections, and arrived at the conclusion that Brandt's "hepatic tissue" was nothing more nor less than a mass of deep-lying epidermal glands. In this he was misled by the circular outline of the botryoidal vessels when in section, and by the fact, which he was the first to observe, that unicellular epidermal glands do travel very deeply into the body-wall of the Leech, and form groups of spherical cells, which are obvious enough in transverse sections (see Mr. Bourne's fig. 13, Plate XXV).

Leydig, in his ' Histology' (1857), states : " Relatively to " the liver of the Leeches, properly so called, my observations " compel me to differ completely from the prevailing opinion. " Certain utriculi, of a brownish-yellow colour, which envelope the " stomach and intestine, are commonly considered to constitute " the liver ; these utricles open one into the other by means of " their excretory canals, and pour their contents on to the internal " surface of the intestine. Contrary to this view, I venture to

"maintain that the tissue of the Leeches, which is regarded as a
"liver, has a very different signification; it ought to be placed
"alongside of the fatty bodies of the Arthropoda. It is formed
"of cells of various sizes and of variable form, round, elongated,
"sometimes drawn out, and fibrous; in other cases the cells are
"ramified and the prolongations anastomose with one another;
"frequently they form tubes with hemispherical prominences; in
"short, they reproduce all the varieties of form which are pre-
"sented by the cells which compose the fatty body of Arthropods.
"In Hirudo, Hæmopis, and Nephelis, the cell-content is formed
"by a brown granular mass in greater or less abundance. Just
"as the fat body of Arthropods is in connection with the external
"membrane of the tracheæ, intestines, &c., so also the tissue of
"the Leeches, which has wrongly passed, up to the present time,
"for the liver, is in connection with the connective-tissue envelope
"of the intestine; it embraces not only the alimentary tract, but
"it also constitutes the brown envelope of the testicular vesicles,
"the *tunica adventitia* of the vascular trunks, the loose brown en-
"velope of the nervous system, &c.; in a word, this liver is simply
"a form of connective tissue, which, in the absence of a proper
"periviceral cavity, fills up all the interstices situated between
"the organs which it invests. Its resemblance to the fat body of
"Arthropods has other facts to support it. Thus, although the
"brown granules make up the bulk of the cells, yet it may be ob-
"served (*Hæmopis*, for instance) that, among the brown-coloured
"networks, there are other fibres, the cells of which have, as con-
"tents, colourless granules of a fatty nature; and, what is still
"more striking, is that in Clepsine and Piscicola a well-developed
"fatty tissue occupies the place of these brown networks. Where
"the cells form, by means of their outgrowths, a system of mesh-
"works the cavities are filled by gelatin. Besides observation on
"fresh specimens, I recommend the following mode of preparation:
" —A leech is thrown into hot water, it is then dried, and fine
"transverse sections are made, which are then soaked in slightly
"acidulated water. It is then clearly seen that the connective
"tissue which envelopes all the organs, taking its point of de-
"parture from the integument, and, traversing all the muscles, is
"filled, in certain parts and in its cellular elements, by brown
"granules, and, moreover, that the colouring matter is of the
"same nature as that of the integument."

Thus, it appears that Leydig had arrived at very nearly the
same conclusion with regard to the relationship of the botry-
oidal tissue and the brown fibrous tissue as that to which I
have been led. He had, however, entirely missed the connec-
tion of both these forms of the vaso-fibrous tissue with the

hæmatophorous vessels, the blood-vessels as they are usually called.

In a much more recent work, however, namely, in his excellent treatise on the anatomy and histology of the Oligochætous worm, *Phreoryctes menkeanus* (' Archiv f. mikrosk. Anatomie,' vol. i, 1865), Leydig has touched upon various points in the anatomy of other worms illustrative of Phreoryctes. He there figures a small piece of the botryoidal tissue of the Leech and remarks on its vascular nature, showing that since the publication of his ' Histologie,' he has ascertained the fact that the cells of the botryoidal tissue form the walls of blood-vessels.

The observations of Leydig which I have above cited relative to the continuity and identity of the epidermic pigment, intermuscular and tunic-forming fibres, and botryoidal or so-called hepatic tissue, show that he had arrived at the conclusion that what I have called the "vaso-fibrous tissue" is one and the same structural element with a variety of modifications, but he has not included in its area the hæmatophorous vessels, as I have found it necessary to do, nor is the very valuable passage which I have cited from the ' Histologie ' explained by drawings either in that work or by those which the author has given in illustration of his most admirable papers, published from time to time in the ' Zeitsch. f. wiss. Zoologie.'

Injection of the vaso-fibrous tissue.—In the course of some experiments made by injecting indigo-carmine beneath the integument of the Leech in order to observe (if possible) the excretion of that substance by the nephridal cells, I was struck by the fact that in transverse sections of leeches so treated, the intermuscular brown fibres of the vaso-fibrous system were very uniformly coloured blue by the indigo-carmine, to the exclusion of other parts of the organism. I am not able to say whether this was a mechanical injection of the tubular fibres, or whether it was rather due to a selective vital action on the part of the vaso-fibrous tissue, similar to that noted in Vertebrate areolar tissue by Arnold. I am inclined to think that it was not mechanical since the vessels of the hæmatophorus system were not injected. This latter fact could, however, be explained on the hypothesis that these vessels were already filled with their hæmoglobinous fluid, whilst the tubular fibres of the brown fibrous tissue were in a non-distended but distensible condition.

On the USE of the WENHAM BINOCULAR with HIGH POWERS. By HENEAGE GIBBES, M.B.

A GREAT many plans have been tried for the purpose of obtaining stereoscopic effect with high powers, and these have all required a special stand or special apparatus entirely removed out of the reach of the ordinary working microscopist, and this led I think to the prevalent idea that the binocular microscope is entirely unsuited for histological or pathological research.

Mr. Stewart, of St. Thomas's Hospital, showed me a $\frac{1}{6}$ of Zeiss's, which, by removing the front part and screwing it on to an adapter, gave perfect stereoscopic effect, and this induced me to try the oil-immersion lenses in the same way. I unscrewed the front of my $\frac{1}{12}$ oil by Zeiss, and screwed it on to an adapter made in such a manner that the lens was brought as close as this form of mount would allow to the prism, and by cutting off the lower part of the slide below the rackwork of the coarse adjustment I was enabled to bring the objective low enough to reach the object.

With this arrangement I found that I could not get perfect stereoscopic effect, but I got it near enough to show me that if I could bring the glass a little nearer the prism the effect would be obtained, the black bands on either side of the field being very small. With this view Messrs. Powell and Lealand made me a $\frac{1}{12}$ oil immersion in which the front is made to screw off just behind the back combination, and a screw is cut on the outside, so that the front can be screwed into an adapter, and this again into the body of the binocular microscope, thus bringing the objective almost in contact with the prism.

With this glass the stereoscopic effect is perfect, the whole field is illuminated, and the result obtained is really wonderful. Taking a preparation of the tadpole's tail hardened in gold solution, the different elements are seen in their true relations to each other; there is no difficulty in deciding whether a fine nerve-termination passes over or under or INTO a connective-tissue corpuscle. Cells are seen not as flat plates, but as spheroidal bodies, with their intranuclear and intracellular network pervading their whole substance.

The only difficulty I have found is to persuade people that the power is really so high, objects stand out in such bold relief, they cannot believe it possible. I have had a $\frac{1}{16}$ oil made by Messrs. Powell and Lealand, and this glass, although

there is a slight shade on either side of the field, gives per-
fect stereoscopic effect.

These glasses are of great excellence, and I think excel
Zeiss's in definition; they have also this advantage, they are
perfectly homogeneous, and require the ordinary cedar oil
for either oblique or central light, and do not require any
adjustment of the draw tube for thin covers, as is necessary
with Zeiss's glasses.

Any ordinary stand made on the Jackson-Lister model can
be used, if the body is made to come down in contact with
the stage, as the objective only projects $\frac{3}{10}$ of an inch.

An achromatic condensor is also necessary.   I have used
Powell and Lealand's and Zeiss's, but any maker's with a
large angle would probably do.

On the STRUCTURE *of the* SPERMATOZOON. By HENEAGE
GIBBES, M.B.

SINCE my last paper appeared in this journal in October,
1879, I have had little time to make further researches into
this subject.

I have, however, examined the spermatozoa of several
animals, such as rat, mouse, axolotl, pigeon, in all of
which I have found the long filament formerly described;
also in the pigeon, where it resembles the Spermatozoon of
an Amphibian on a smaller scale.

I have examined a few Invertebrates, but so far have only
found the filament in the snail and leech. In the snail
it appears to be only a little longer than the tail; this,
however, is enormously long, which may account for it.

In the fowl the Spermatozoa resemble that of the newt in
shape, but the filament is very fine, and consequently indis-
tinct. I have examined a few fish, but the spermatozoa were so
small I could not make out anything satisfactorily.   I have
examined a number of specimens of the human spermatozoa,
some taken from the testes twenty-four hours after death by
accident, others from twelve to twenty-four hours after coi-
tion; these came by post, and I could detect no material
difference in their structure, with the exception of the vary-
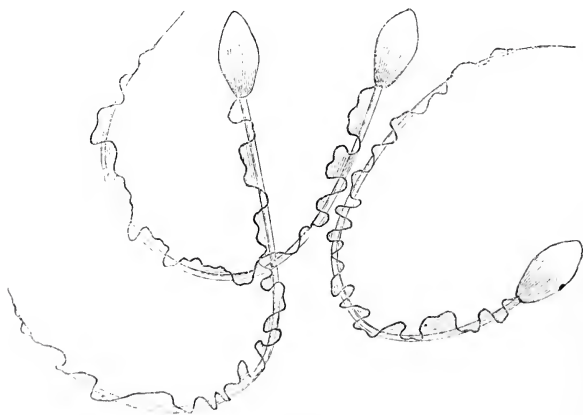
Human Spermatozoa.

ing lengths of the tails in different specimens.   I think
something may be made of this, if one could only get suffi-

cient data to go upon. In all I found the filament very fine, and requiring great care in the illumination to show it properly, as when, after a good deal of trouble it was shown well with the $\frac{1}{25}$ immersion, I found I could readily see it with the $\frac{1}{16}$ oil, and even the $\frac{1}{12}$ oil immersion.

The drawing was made from a specimen prepared in a mixture of glycerine and absolute alcohol, and drawn with Messrs. Powell and Lealand's $\frac{1}{25}$ oil immersion and A eyepiece. The filament is very fine, and is connected to the tail by a membrane, which is much wider than in the Amphibia, and allows it to move further from the tail; it is also longer than in the Amphibia, and is more folded in consequence. I found in one specimen a number of heads with no corresponding tails. I have not, however, been able to get at the history of the individual who supplied it.

Some disputed points in ECHINODERM MORPHOLOGY. By
  P. HERBERT CARPENTER, M.A., Assistant Master at
  Eton College.

THE last number of the ' Zeitschrift für wissenschaftliche
Zoologie ' (Band xxxiv, Heft 2) contains two valuable papers[1]
on Echinoderms, by my friend Dr. H. Ludwig, of Bremen.
Many new observations of the highest interest are here
recorded, and it is not too much to say that their accuracy
may be relied on with the utmost confidence. But, at the
same time, I believe that some of the conclusions which
Ludwig has drawn from them are essentially unsound; and
in the following pages I propose to state my reasons for this
belief.

In the first place, however, I desire to state that some of
the observations recorded by Ludwig in these papers have
convinced me that certain views which I have advocated
in the pages of this journal are no longer tenable. Four
years ago Ludwig described some tubular appendages of the
water-vascular ring of the Crinoids as opening below into
the body cavity into which they depend ; and he therefore
considered them together with the water-pores of the disc as
collectively representing the madreporic apparatus and sand
canals of the other Echinoderms.

Like Greeff, however, I did not feel quite satisfied respect-
ing the alleged opening of these tubes into the body cavity,
as I knew from my own observations of them (made like
those of Ludwig by the section-method) that the liability to
error was considerable. But now that Ludwig has found
them to be open in the uncut Pentacrinoid larva I see no
reason to doubt that the same is the case in the adult.
Consequently, the resemblance[2] that I, like Professor
Huxley, believed to exist between these (apparently cæcal)
tubes and the closed *vasa ambulacralia cavi* of the Ophiurids
will not bear investigation.

But, while abandoning this position, I must still confess to
a lingering doubt as to whether the disconnected water-pore
and water-tube of the *Antedon* larva can be regarded as
perfectly homologous with the madreporic system of the
other Echinoderms. I fully admit the similarity of their

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[1] "Ueber den primären Steincanal der Crinoideen nebst vergleichend
anatomischen Bemerkungen über die Echinodermen überhaupt," pp. 310—
332, Taf. xii u. xiii.   "Neue Beiträge zur Anatomie der Ophiuren," pp.
333—365, Taf. xiv—xvi.

[2] This Journal, vol. xix, New Ser., p. 10.

histological structure and of their functions as afferent channels of the water-vascular system. But this similarity does not constitute an homology. The dorsal pore, which is developed so early in the other Echinoderm larvæ as the foundation of the madreporic system, is absent from the corresponding stages of the *Antedon* larvæ;[1] and until the development of the primary water-pore and water-tube of the Crinoids are more fully known, I do not think that we can without qualification accept Ludwig's view regarding their homology with the madreporic system of other Echinoderms. The following remarks, however, are based upon the assumption of the truth of that view.

Ludwig regards the external opening (water-pore) of the madreporic canal (water-tube) as a fixed point. Whether it be actinal or whether it be abactinal does not matter. In the larvæ of some Crinoids and in the adult Ophiurids it opens on one of the interradial actinal plates (orals) around the mouth. In the adult Urchins and Starfishes, however, it opens on one of the interradial abactinal plates around the anus (genitals). This is far from being the case in young forms of either class. Nevertheless, Ludwig thinks the relations of the parts in the adult sufficiently fixed to enable him to assert the *undoubted homology* of the orals of Crinoids and Ophiurids with the genitals of Urchins and Starfishes. He attempts to strengthen this remarkable position by other considerations to which I shall presently advert; and he points out how diametrically opposed it is to the view now generally held that the *Basals* of Crinoids represent the genitals of Urchins and Starfishes.[2] He seems, however, not to attach much value to the evidence on which

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[1] I have often wondered whether the " pseudoproct " of Wyv. Thomson, a depression at the hinder end of the very young *Antedon* larvæ, can be regarded as representing an undeveloped dorsal pore. Its position at the end of the transverse Gastrula-axis is the same as that of the dorsal pore in other Echinoderm larvæ, though I am bound to admit that it is not dorsal. But owing to the extreme elongation of the Gastrula along that axis it is very far from the water-vascular ring and not close to it as in other larvæ. This might account for its want of further development. On the other hand, the primary water-pore of the *Antedon* larvæ leads into the oral cœlom developed from the left peritoneal vesicle of the embryo. It is not likely, therefore, ever to have been a simple *dorsal* pore, or it would have opened into the aboral cœlom developed on the dorsal side of the larva from the right peritoneal vesicle, supposing of course that it were not directly connected with the water-vascular ring by a water-tube, as in other Echinoderms.

[2] See " The Oral and Apical Systems of Echinoderms," this Journal, New Ser., vols. xviii and xix ; and also the recently published text-books of Claus, Zittel, and other authors.

this view is based, or he would scarcely have written as follows (p. 317) :

"Der ganze Beweis für diese Auffassung liegt darin, dass, wenn man von dem Mittelpunkt der dorsalen Oberfläche des Thieres ausgeht, bei den Crinoideen die Basalia, bei den Echinoideen die Genitalia die ersten Platten sind, welche stets und immer in der Richtung der Interradien angeordnet sind. Irgend welche unmittelbaren Beziehungen zu den inneren Organen des Thierkörpers sind bei dieser lediglich auf die räumliche Auordnungsweise jener Platten gegründeten Homologisirung nicht in Betracht gezogen worden."

The view attacked by Ludwig rests, however, on a much firmer foundation than the mere anatomical fact which he calls the "ganze Beweis" of its truth. The evidence of Echinoderm embryology is all in its favour; but Ludwig does not make the slightest mention of this evidence, much less attempt to controvert it.

It is as follows :—The interradial abactinal plates (basals) of Crinoids are developed with precisely the same relation to the vaso-peritoneal apparatus of the larva as are the inter-radial abactinal plates (genitals) of Urchins and Starfishes. In both cases these plates first appear in the form of a spiral around the right peritoneal vesicle (Agassiz, Götte); but the orals of the Crinoids, which Ludwig considers homologous with the genitals of Urchins and Starfishes, are developed spirally round the left peritoneal vesicle (Götte). Are not these peritoneal vesicles as "important inner organs" as any in the whole morphology of the Echinoderms? They develop before the water-vascular apparatus, on the connection of which with certain plates of the adult, Ludwig lays such stress.

He admits that the oral or actinal side is homologous in all Echinoderms as being chiefly developed from the left side of the larva; and yet he considers plates developed on the left or actinal side of one Pluteus larva (Ophiurids) as undoubtedly homologous with those developed on the right or abactinal side of another (Echinids), and leaves the abactinal plates of the Ophiurids (genitals) out of consideration altogether.

It seems to me that he assumes too much in regarding the water-pore of the adult as a fixed point. If he could show that one of the genitals of an Urchin or Starfish larva were primitively perforated by the water-pore as is one of the orals of a larval Crinoid, then his position would be strong indeed. But this is very far from being the case. He admits himself that in young Starfishes (Lovén) there is no connection between the water-tube and any genital plate.

The observations of Metschnikoff and Agassiz are quite in accordance with this fact. The former figures the apex of a young Starfish in which the madreporic plate is already formed, but is situated at the edge of the disc quite outside the circle of genital plates; while Agassiz represents a young Starfish which "shows the position of the madreporic body immediately on the edge of the disc of the lower (*i.e.* actinal) side." Somewhat the same is the case in the adult Ophiurids. Although developed on the dorsal surface of the larva the water-pore is usually on the actinal surface of the adult; while in *Trichaster* it is neither abactinal nor actinal, but intermediate in position, somewhat as in the earlier stages of the young Starfish.

In the same way the anus of the Starfish is at first "upon the actinal side near the edge of the disc." But as growth proceeds it moves towards the abactinal surface together with the water-pore. In the Crinoids, however, the position of the future anus is gradually shifted in the reverse direction, *i.e.* towards the actinal surface; while the water-pore must be developed late, unless we are to suppose that it escaped the notice of both Götte and of Wyville Thomson in the earliest larval stages. But whereas in the other Echinoderms it is developed very early, before the appearance of the actinal or abactinal plates, the rudiments both of these and of the chief organs of the Crinoid appear before it, and then it is only found perforating the "seitlichen Randtheil" of an oral plate.

As far as the Crinoids are concerned, this is the chief evidence in favour of Ludwig's views, but it is not complete even for this group. For while *Rhizocrinus* has five water-pores like some exocyclic Urchins, their openings are not in the persistent oral plates[1] as they should be on Ludwig's theory, which compares these orals to the genitals of the Urchins, because one of them in the larval *Antedon* is perforated by the water-pore. Further, the orals of the Crinoids have precisely the same relation to the water-vascular ring and to its tentacular apparatus surrounding the mouth as the five plates figured by Kowalevsky in the *Psolinus* larva. Ludwig will hardly deny that these last are orals, but not one of them is perforated by the water-pore. On the contrary, it is separated from them by the whole length of the

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[1] If the orals of *Rhizocrinus* are perforated by the water-pores, surely Ludwig would have said so; and I gather from his figure of the water-pore in this genus that this is not the case ('Zeitschr. f. wiss. Zool.,' xxix, Taf. v, fig. 8).

water-tube, which extends backwards from the water-vascular ring and not forwards as it should on Ludwig's theory.

Again, what is the position of the five water-pores of *Trichaster elegans* ? Not close round the mouth in the position of the absent mouth-shields, but between the two genital clefts of each interradius. The interradial plates sometimes developed here in other Ophiurids have been hitherto regarded as representing the genitals of Starfishes and Urchins; and the presence of the water-pores in the same position in *Trichaster elegans* goes a long way towards strengthening that view, while at the same time it diminishes the probative value which Ludwig assigns to the perforation of the mouth-shields by the water-pores in other Ophiurids.

Besides the perforation of one of the orals of the larval Crinoid by the water-pore, the only other arguments adduced by Ludwig as showing the homology of these orals (*vice* the basals, discarded) with the genitals of Urchins are the following :—(1) The anus is adoral from the basals of a Crinoid, but aboral from the genitals of a regular Urchin; though it is aboral from the orals of a Crinoid. I would urge two considerations in reply. There *are* Crinoids in which the mouth is at the margin of the disc, close down to the abactinal skeleton, while the anus is central; and I do not see why these and the irregular Urchins should be left out of consideration, especially when we remember that the mouth of the Pentacrinoid larva is excentric, as Ludwig himself has told us, and that the exocyclic Crinoids present other embryonic features.

In the young Starfish the anus is primitively adoral from the genital plates, and only secondarily assumes the intragenital position on which Ludwig bases his argument. Here, as in other cases, Ludwig's homologies seem to me to rest too much on the variable secondary relations of organs as existing in the adult, and too little on the constant and primary relations of their rudiments in the larva.

(2) Ludwig lays great stress on the homology of the oral side in all Echinoderms, as shown in the following interesting facts. When viewed from the oral side the gut always winds from mouth to anus in the direction taken by the hands of a watch, and the water-pore both of the Urchin and of the embryo Crinoid is in the same interradius as the foregut. He gives an excellent figure (Tab. XIII, fig. 7) of the course of the gut in an Urchin as seen from the ventral side, and therefore (of course) represents the madreporite as seen from within in the N.W. corner of the figure. He gives a similar figure of the course of the gut in *Antedon*

(fig. 9), but without representing the " homologous " oral plate with its water-pore. Had he done so he would have been obliged to explain that while in the Urchin figure the madreporite was represented as seen through the gut, in the Crinoid figure the reverse would be the case, viz. the gut seen through the perforated oral plate. But nevertheless the latter is homologous with the genitals of an Urchin!

Again, the positions of the primary actinal and abactinal plates, with respect to the oral centre, is not the same in the Urchins and Crinoids if the latter be " orientirt " according to Ludwig's theory. In an Urchin the proximal plates to the oral centre are the oculars (radial), and the distal series the genitals (interradial).

In a Crinoid or Ophiurid the interradial orals are proximal and the radials distal; but, nevertheless, the homology of the orals and genitals is undoubted, while the interradial basals of the Crinoids, which are beyond the radials just as the genitals of an Urchin are beyond the oculars, are not represented in an Urchin! Further, Ludwig takes no account of the five interradial plates which surround the mouth of *Palæostoma*, and have the same relation to the water-vascular ring, &c., as the orals of *Rhizocrinus*, a relation which, it is almost needless to say, is *not* characteristic of the genitals of an Urchin.

(3) Another of Ludwig's arguments is founded upon the peculiar relation of the aboral blood-vascular ring of the Ophiurids to the mouth-shields. Because it dips down in the five interradial spaces, and partially rests upon these plates, Ludwig considers them homologous with the genitals of the Urchins and Starfishes. But the rest of this blood-vascular ring remains in the abactinal position that the whole of it occupies in the Urchins and Starfishes, and it is from this abactinal position that the principal vessels are given off to the genital glands. According to Ludwig (p. 352), " Dieses Genitalgefäss entspricht sowohl in seinem Ursprunge als auch in der Richtung seines Verlaufes und in seiner Beziehung zu den Geschlechtsorganen den Genitalgefäss der Asterien." On Ludwig's theory, however, one would expect the genital vessels to be given off from those parts of the blood-vascular ring which lie nearest the ventral surface, since it is in the plates of that ventral surface that he recognises the homologues of the genital plates of the Starfishes. One would also expect that these ventral plates would be occasionally perforated by the openings of the genital glands, But is this ever the case? Ludwig

admits that it is not ; and the mere fact that in some Urchins and Starfishes the genital ducts do not open on the genital plates does not seem to me a sufficient argument to meet this difficulty, though, according to him, " Indessen stört das die Homologie, die nach Obigem zweifellos *zwischen den Mundschildern der Ophiuren und den Genitalplatten der Echinoïdeen und Asterien besteht durchaus nicht* " (! ! !)

Ludwig nowhere says a word about the genital plates of the Ophiurids, and yet these are developed interradially on the abactinal surface of the larva precisely like the genital plates of the Starfishes. The following passage from Agassiz ('North American Starfishes,' p. 93) is interesting as regards this question :

"In Ophiurans the genital plates are formed from the angles of the five interradial plates ; similar plates can still be traced in the young Starfishes, while in the full-grown Starfishes their presence is shown by the interbrachial partition, on each side of which the ovaries discharge. Thus there exists a complete homology between the genital plates of Ophiurans and the interbrachial partitions of Starfishes, a homology fully carried out in its details when we examine the relations held by the genital plates to the ovaries in Ophiurans and by the interbrachial partitions to the ovarian openings in Starfishes."

Of course there are aberrant forms with more or less exceptional peculiarities among the members of each class, but the general relations of both classes are such as entirely to support the statements quoted above, while the peculiar features of the exceptions (*Asterina gibbosa, Trichaster elegans*, &c., as shown by Ludwig's own observations) seem to me to be of such a nature that they support Agassiz's view rather than Ludwig's very revolutionary one.

The relations of the primary actinal and abactinal plates to the blood-vascular system may be looked at from another point of view than the one used by Ludwig. A person standing in the central plexus (heart) of a young Starfish, with his feet in the oral blood-vascular ring, would have the genital plates above his head. But if he were similarly placed in a Crinoid (with the ventral side downwards) he would have the basals over his head, and not the orals, although these last are the homologues of the genitals of the Starfish according to Ludwig's theory.

Ludwig does not attempt to follow out to any great extent the logical results of his theory with respect to the relations of the various regions of the body among the different Echinoderms. But he points out (p. 319) that they involve our believing in a great difference between Urchins and

Crinoids, one, namely, which has not been thought to exist since the researches of Agassiz and Lovén. While the Crinoid calyx increases by the additions of plates on the aboral side of the oral plates that of an Urchin grows very differently:

"Hier entfernen sich die Oralplatten der Crinoideen homologen Genital-platten immer weiter von dem Munde, indem die Bildung des Perisoms des erwachsenen Thieres, genauer die Bildung der Interambulacralfelder, an der adoralen seite der Genitalplatten erfolgt."

This must not be understood to mean (as it well might do) that the genital plates have the same primitive relation to the mouth of an Urchin as the orals have to the mouth of a Crinoid and only subsequently become separated from it, for this is far from being the case.

The old view that the genitals of an Urchin represent the Crinoid basals involves none of the difficulties inseparable from Ludwig's new one. In both Crinoids and Urchins[1] new plates are added on the adoral side of the interradial abactinal plates, viz. the genitals of the latter and the basals of the former, which Ludwig, however, considers to be unrepresented in the Urchins.

The only other out of the "Reihe wichtiger Folgerungen für die vergleichend-anatomische Auffassung der Skeletregionen der verschiedenen Echinodermgruppen" that Ludwig deduces from the homologies which he asserts is the following[2]:—"Dass das perianale Feld der Echiniden dem gesammten Perisom der Ophiurensheibe mit Ausnahme der Arme und der Mundschilder homolog ist."

There seems to me to be something wrong here. Perhaps it is only that Ludwig and I understand the expression "perianale Feld" in different ways. I take it to mean the ring of genital and ocular plates, and, as Ludwig considers the former to be homologous with the mouth-shields of the Ophiurids, I am somewhat puzzled as to the meaning of the above passage.

The above are many inconsistencies which become apparent upon a critical examination of this new theory of Echinoderm morphology. It is difficult to understand how Ludwig can insist on the homology of the oral surfaces of Urchins, Starfishes, and Ophiurids, and yet insist even more strongly that plates around the mouth of the latter are homologous with plates around the anus on the aboral

[1] This Journal, New Series, vol. xix, pp. 26—28.
[2] 'Zoologischer Anzeiger,' 1879, ii, p. 542.

surface of the two former groups.   His theory reminds one
of Kowalevsky's celebrated attempt to prove that the brain
of a worm represents the hinder end of the Vertebrate spinal
cord; and one cannot help feeling that, despite the un-
doubted value of his observations, some (though by no
means all) of the conclusions which he has based upon them
will not bear a close investigation when compared with
the primary facts of Echinoderm structure.

The ORIGIN *of the* RED BLOOD-CORPUSCLES.　By Professor
POUCHET, of the Jardin des Plantes, Paris.[1]

I HAVE paid a good deal of attention lately (since November,
1877) to the study of the formed elements of the blood and
their origin.　The fact was, as often happens, that several
histologists were pursuing, both in France and elsewhere, the
same investigation.

I propose to give a summary of what seems to me to be
the actual state of our knowledge on a subject of such
importance both to physiology and to medicine.

A sufficiently large part in these studies, which have been
followed out with vigour in various quarters, has been given
to the history of that singular phenomenon the coagu-
lation of the blood.　I shall put this subject entirely on one
side, only wishing to occupy myself here with the formed
elements of the blood in the living state, with their origin
and the phases of their existence, in so far as they are con-
stituent elements of the organism.　Coagulation is a post-
mortem phenomenon.

I shall speak in the first person, giving credit where
credit is due to others as well as to myself.

It is not a little peculiar that the formed elements of the
blood, which have been so long known, should be so little
known.

Even to-day we cannot say that we know with absolute
certainty how they originate (at least in the adult), how long
they live, and what may be their fate, for it is certain that
their existence is relatively short.

Every day one sees in the hospitals patients losing large
quantities of blood, and it seems as though no one had asked, or
at least had seriously considered and sought to discover, how
it is that, at the end of a few weeks, the blood lost is entirely
replaced.

G. E. Rindfleisch, in calculating the replacement of blood
in women during the intermenstrual period, estimated that
half a centigramme must be produced in a minute, which
means that about 175 millions of hæmatids are produced in
the body every minute.[2]

How does this enormous proliferation come about ?　It is
not a little hard to be obliged to confess that we are reduced

[1] Translated from the 'Revue Scientifique.'
[2] The term 'hæmatid' is a rendering of the French term 'hématie,' which
M. Pouchet uses to denote red corpuscle.

to hypotheses. It is upon this obscure problem that I wish to throw some light.

One need only look back over the last two years of the morphological study of the blood[1] to see how many organs have been put forward, one after another, as the birthplaces of the corpuscles, both white and red.

Theories and hypotheses have not been wanting; the lymphatic glands, the spleen, the marrow of the bones, and other organs, have been looked upon as the seat of origin of the formed elements of the blood, and even called, on that account, " hæmatopoietic organs." It must be allowed that the question of hæmatogenesis is far from being a simple one, and that among various animals it presents itself under very different aspects.

We may recognise, in the first place, two great classes:

Firstly.—Oviparous animals, either warm- or cold-blooded, in which the hæmatids possess a nucleus.

Secondly.—Mammals, in which the hæmatids, although at first like those of oviparous animals, are replaced at a very early stage by corpuscles, absolutely devoid of a nucleus.

If one considers all animals with regard to their hæmatopoietic organs, it is evident that many animals have neither marrow nor lymphatic glands, e. g. fishes.

In the same way there are some in which the spleen is quite rudimentary (Syngnathus), or even entirely wanting (Lampreys).

Among mammals, even the Rodents, in which the vascular area persists and forms blood-corpuscles at a very late period ; and, on the other hand, the Marsupials, in which the umbilical vesicle has disappeared, even before any part of the primordial skeleton has become vascular, offer various points which call for special attention.

In order to solve the problem of hæmatogenesis, we want to obtain a large number of data which would be useful to us.

By what certain characteristics can we recognise those elements which are newly formed from those which are about to degenerate? How long do they live? For it is impossible to admit the long life of elements, the regeneration of which is so easy after lesions, and even in the normal state of things in woman between puberty and the critical age.

The more or less satisfactory answers to all these questions, which have been made by recent research, I wish to

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[1] 'Gazette Médicale,' Nov. 10th, 1877; Jan. 19th, Feb. 2nd, March 16th, April 27th, 1878; and 'Journal de l'Anatomie,' Jan., 1879.

present here in an order which, if not the most logical, is probably the best for a rapid *résumé*.

It is curious that most of those who have worked at hæmatogenesis have been led to recognise from the first the inaccuracy and incompleteness of the descriptions which have been given, up to the present, of the formed elements of the blood. One would have thought that objects so easily examined would have been well known. Nothing of the sort. Certain elements, of perhaps capital importance, have been completely neglected, others badly described, while as to the origin of all, one is reduced to the vaguest hypotheses.

In short, our first want seems to be to complete here, as well for the oviparous vertebrates as for the mammals, such descriptions as are found in the recognised text-books, and even in a whole host of special memoirs on the subject, which have been recently published.

Blood of Oviparous Animals.—Among all oviparous animals—those which are warm-blooded—birds, as well as those which are cold-blooded—reptiles, amphibians, and fishes—the method of formation in the adult seems to be the same. Among embryos it evidently differs according as the embryo has or has not an umbilical vesicle. In the adult the question of the formation of the elements of the blood seemed actually settled. The size of the elements in Amphibia naturally induces histologists to study these first. I took the newt as the basis of my investigations.

There are in the blood of the newt certain elements not corpuscles, which have hitherto all been included under the general term leucocytes. These I have called, for the sake of clearness, "nuclei of origin;" however, by no means insisting upon this term. These "nuclei of origin" are spherical, and about $\frac{10}{1000}$ mm. to $\frac{12}{1000}$ mm. diameter. They are enveloped by a very thin layer of protoplasm, of equal thickness all round the nucleus. I will return presently to the origin of these elements ; it is their further development which concerns me at present. This development may take place in two different methods :

a. The " nucleus of origin" enlarges and segments, and the protoplasm becomes proportionally increased in bulk ; in short, the nucleus becomes a leucocyte with numerous nuclei. After this stage, when it may be called adult, it must necessarily disappear ; the protoplasm disintegrates in the serum, setting free the nuclei, which are nothing else than nuclei of origin about to recommence a new cycle. This appears to be the normal state of things.

b. The nucleus of origin may be destined to become a

hæmatid, to this end undergoing a sort of abortion or degeneration.

The protoplasm now commences to form hæmoglobin ; the presence of the latter seems to be shown from the first by the shape which the protoplasm assumes, which is already a geometrical one.

The nucleus does not multiply; the element which has thus undergone hæmoglobic degeneration has reached by the process its ultimate form, and is destined to disappear more or less slowly.   The nucleus, as it takes on an oval shape, in accordance with the elongated shape of the protoplasm, seems to lose that power of dividing and multiplying which it originally possessed.   The protoplasm increases and commences to take on a yellow colour, visible with a microscope, on account of a continued accumulation of hæmoglobin.

Soon it has entirely lost its essential vital properties, sensation, and power of movement.   It becomes, with its nucleus, a passive body like the cells of the horny layer of the epidermis, retaining its chemical affinities, but devoting itself entirely to this chemical function of hæmoglobic degeneration.   Hæmoglobin continues to accumulate in the protoplasm, which thus becomes more and more dense and coloured.

The nucleus loses its chemical characters and the protoplasm soon constitutes a homogeneous mass, which is finally dissolved in the blood plasma ; it may be while still in the circulation; it may be, on the other hand, after having been stopped on account of its diminished elasticity in the parenchymatous tissue of the spleen.

I believe I was the first to completely describe the successive stages of this development, as well as a certain number of points of less importance which go along with it, but on which it is useless to dwell in this rapid *résumé*.

M. Hayem has called the young corpuscles, the bodies of which are already flattened, oval, and still appear colourless, it may be, on account of the extent to which they are magnified, hæmatoblasts.

It seems certain that these young corpuscles are at least capable of changing their shape in the blood in a state of rest, as do the nuclei of origin and the adult leucocytes.

M. Hayem admits the transformation of his hæmatoblasts into corpuscles.

But M. Vulpian ('Comptes Rendus,' 4th June, 1877) certainly has the credit of having demonstrated this development; he has shown by careful experiments that in the blood of dead frogs hæmatoblasts always appear in large numbers,

and gradually become transformed into red corpuscles (hæmatids); he gives a very accurate description of these hæmatoblasts. Repeated experiments on birds have afforded me precisely the same results. Neither M. Vulpian nor M. Hayem have put forward any opinion as to the origin of these hæmatoblasts. I have stated before that they are derived from nuclei of origin. But whence do these latter come? Do they all come from the broken and scattered nuclei of leucocytes which have finished their existence?

Fishes have no lymphatic glands. Some animals have no spleen. Is it possible to admit that the nuclei of origin originate, in this latter case, from the cells lining the walls of the lymphatic cavities, and that in the other oviparous animals the spleen, if not the exclusive place of their production, at any rate plays an important part in it? In fact, I have shown that among Selachians the spleen is partly formed of elements which are identical with these nuclei of origin, and which, detaching themselves one by one, fall into the network forming the tissue of the organ, and are carried away by the blood. It must be admitted that the spleen plays, among oviparous animals, a part which it plays, perhaps, among the mammals, and which corresponds, at all events, to that of the lymphatic glands of the latter group.

Lastly, it results from certain experiments which I performed some time ago upon birds, Amphibia, and fishes, that the removal of the spleen only hinders the reparation of the blood after death, and that either the nuclei of origin are continually originating from the walls of the lymphatics, or that the leucocytes of the blood are sufficient for this multiplication.

Leucocytes of Semmer.—When the blood of an oviparous animal, and often when that of a mammal, is examined, it is possible to find certain elements, the nature of which has been for a long time misunderstood.

I thought I had been the first to describe them in the blood of Selachians (Soc. de Biologie, Nov. 7). I have since remembered that attention had already been drawn to them in a special manner among mammals by a pupil of Alexander Schmidt, in a paper written at Dorpat. I propose to call them, after the author of this paper, leucocytes of Semmer.

Even if their existence was known, their fundamental structure had escaped all previous observers; they may be regarded as leucocytes (they are the same size, possess amœboid powers, &c.), in the body of which hæmoglobin is formed. The latter is in large granules, generally measuring

from $\frac{1}{1000}$ to $\frac{1\cdot5}{1000}$ mm. They possess all the physico-chemical characters of the hæmatids.

We are now able to understand the full importance of this discovery, which, perhaps, partially escaped M. Semmer. It shows us that hæmoglobin can exist in other anatomical elements than in red corpuscles. M. Kühne, before this, thought that the colouring matter of the red muscles of vertebrates might be hæmoglobin, but he was not able to prove it. Now, it is easy to prove by all the usual tests that the granules in the leucocytes of Semmer are made of hæmoglobin. If MM. Alexander Schmidt and Semmer have described them as an intermediate form between leucocytes and red corpuscles, this must not be understood in the sense that they represent a developmental stage through which the leucocytes pass before developing into red corpuscles: the leucocytes of Semmer are intermediate between the white and red corpuscles only in the sense that they partake in a certain manner of the constitution of both.

Probably the leucocytes of Semmer undergo a final disintegration, and the large granules becoming dispersed in the blood plasma soon dissolve.

Their nuclei are always or almost always near the surface of the cell (which is perhaps important with regard to their special development), and never present any more than do those of other white corpuscles—any trace of decay, and probably become nuclei of origin.

Nothing has hitherto gone to prove that these, originating from a cellular body where hæmoglobin is already formed, can be specially considered to become hæmatoblasts.

First Red Corpuscles of the Embryo.—The formation of the first red corpuscles in the vascular area of birds has been studied by many embryologists, and still raises a certain number of important questions on which there are two opinions. Among oviparous holoblastic animals—the Amphibia—it appears certain that the embryonic cells are directly transformed into red corpuscles, as they show those vitellin grains and small pigment granules which are seen in the corpuscles in their early life, and which only very gradually disappear.

Blood of Mammals.—In spite of the numerous works which have been lately published on the number of the corpuscles (Manassein Malassez, Hayem, Perier, &c.), it is evident that the descriptions given of the elements of the blood are incomplete, and even inaccurate; the characters of the white corpuscles badly defined, the form of the red corpuscles insufficiently described, other elements altogether

neglected. It is, therefore, necessary before going further to go over these various points.

White Corpuscles.—I believe I was the first to describe that the leucocytes of man and other mammals have always, when completely developed, four nuclei, regularly arranged, grouped in the centre of the protoplasm, and devoid of nucleoli. This form represents the adult state. There are always found besides these other smaller corpuscles, few in number, in the blood, in great abundance in the lymph. These have a single nucleus furnished with nucleolus and surrounded by a much reduced layer of protoplasm. These corpuscles are true nuclei of origin, and represent the young state of the quadri-nucleated corpuscles.

They originate in the lymphatic glands, and perhaps also partly in the spleen (see below). What becomes of these quadri-nucleated corpuscles ? The constant production of corpuscles in the lymphatics, the absence, or at any rate the invisibility, of any nucleolus in the four nuclei of the adult corpuscle, leave little room for the hypothesis that these nuclei outlive their protoplasm, and becoming free remake nuclei of origin. It seems equally improbable that the adult quadri-nucleated corpuscles leave the vessels, as has been supposed, to become the differentiated elements of more or fewer of the tissues. In any case, the first thing to do to support this hypothesis, would be to search in the developing tissues for these corpuscles, which are easily recognisable on account of their four, regularly disposed, nuclei.

The leucocytes of Semmer are to be found among mammals as among oviparous animals. They are particularly abundant in the horse where M. Semmer has chiefly studied them.

Red Corpuscles.—I was the first to show that all the red corpuscles of mammals have not necessarily—apart from accidental injury—the discoidal shape, commonly described and figured. Besides the classical red corpuscle, there are always others ovoidal or even fusiform in shape. Their long diameter is longer than the diameter of the normal corpuscle, their edges are slightly turned up. Sometimes these corpuscles are even more elongated, terminating it may be in a point at each end, as in the rat.

It must be noted that this is in no way a question of accidental stretching. I have directly proved its existence in these hæmatids in the blood in circulation in mammals.

We shall see directly the importance as regards hæmatogenesis of this form of hæmatid, which has hitherto only been

noticed in the group of camels[1] (Camelus and Auchenia), where it appears to be universal.

Globulets.—There is in the blood a third kind of element, which anatomists as well as medical men have almost completely ignored, and to which M. Hayem has lately suddenly drawn attention. These elements were discovered by Donné in 1838, and it seems only right to preserve for them the name[2] of the French microscopist who discovered them. In 1846, a German doctor, Zimmerman, blames his fellow countrymen for forgetting in descriptions of the blood, the globulets of Donné, and proposes to call them elementary corpuscles (elementare Körperchen). Although described again by M. Robin, they have remained almost altogether ignored, or else confounded with the various amorphic granules always to be found in the blood.

M. Hayem has given to these bodies the name hæmatoblasts, and has since described them in rather a summary manner, omitting certain points which seem extremely important. I believe I was the first to give an exact description of them.

These bodies are really chiefly remarkable for the precise morphological character which distinguishes them from simple granulations. Zimmerman was not after all so much mistaken about this as Donné and others; he called them corpuscles.

They are elongated, and, except in the very smallest, one diameter is much longer than the other. They are slightly refractive, they appear homogeneous, they are devoid of nucleus, and are unacted upon by staining reagents (?). On the contrary, they approach in their physico-chemical characters the substance of the body of leucocytes. This analogy is even more striking.[3]

[1] Upon what biological peculiarity does the form of the hæmatids in these animals depend? The problem appears to be hard to resolve in the present state of science. The ancestors of this group of mammals appear to be indigenous in regions of the globe, such as the Andes and the plain of Central Asia, where the depression of the barometer is considerable. This form of hæmatid appears to us to indicate a special property of hæmoglobin.

Among fishes, the Syngnathians have regularly nucleated discoidal hæmatids like those of mammals, whilst other Lophobranchs have them extremely elongated and fusiform.

G. E. Rindfleisch has recently stated that the form of the hæmatids is only the consequence of their rubbing one against the other in the serum; the context is sufficient to show what this singular hypothesis is worth.

[2] The French word, used by Donné, was not 'globulet,' but 'globulin.' —TRANSL.

[3] This analogy would lead one to suppose that the globulets were derived from the body of the leucocytes. One might bring forward to defend

They possess an extremely marked tendency to run together, either with one another, with the leucocytes, or with the hæmatids, even in the blood in circulation when under abnormal conditions.

The proportional number of the globulets varies considerably according to circumstances. They are exceedingly easy to study in very young cats; speaking generally, they are not easily seen in animals whose blood is being repaired.

Length of duration and fate of the hæmatids.—It is quite clear that parts of our organism which are as much exposed to accidental losses as are the elements of the blood, and even to periodic losses which accompany certain functions, ought to be always undergoing a regeneration, this regeneration only becoming more or less active according to circumstances.

A consequence of this constant regeneration is that, although one must consider some element in the body as perennial, *e. g.* nerve cells, it is hardly possible to think so of either hæmatids or leucocytes. The elements of the blood have therefore a short existence, how short we know not; various things point to its being only a few weeks, or at most a few months.

If mammal's blood is transfused into bird's blood, the hæmatids are found unaltered at the end of fifteen or twenty days.

Brown-Séquard found this to be the case, and I have since repeated the experiments, using the dog and pigeon. On the other hand, the hæmatid of birds placed in a mammal are not to be found after a few hours only ; this may simply be attributed to the much greater diameter of the hæmatid of the bird. These latter seem to be almost immediately killed and destroyed by the state of the new medium in which they are placed.

If it is difficult to ascertain the absolute age of a hæmatid, it seems easier to determine their relative age.

As a rule with mammal's hæmatids as well as with those of oviparous animals, the nearer they are to their period of decline and disappearance the more coloured and refractive are they. There is no doubt that the hæmatids end by becoming dissolved in the serum. They diminish in volume, and finally take a more or less regular spherical form. They answer in this state to the description of elements found in

this hypothesis a more or less distant analogy, between this phenomenon and the formation of the directive corpuscles ; a common character being the regular segmentation of the nucleus of the leucocytes into four and the regular segmentation of the vitellus, &c.

abundance in the blood in certain diseases by MM. Vanlair and Masius, and which they called *microcytes*.

These old hæmatids have the same general characters as those of oviparous animals. They lose in great part their elasticity, and seem to have on account of this a great tendency to be caught, and remain in the reticulum of the spleen where the blood coming from the arterial extremities falls into this spongy network, out of which open the large venous trunks. There is nothing to support the theory that an active destruction of blood-corpuscles goes on in any special organ.

Origin of Blood in Mammals.—In mammals, as well as in birds, the first corpuscles are formed in the vascular area. I have been able to follow this formation in the rabbit, and to prove that hæmatids continue to be formed from the old walls of the umbilical vesicle, even when the embryo is as much as 22 mm. long.

If the chorion be removed from the surface of the egg the vascular area is exposed, this consists of a layer of similar cells. I have assured myself that the primitive or blastodermic cells differentiate in two ways, forming on the one hand the endothelial cells of the vascular walls, and on the other, the embryonic red corpuscles. The former envelop the latter, which latter, grouped in cæca opening on to the spaces already traversed by the blood, undergo a more or less complete development before being in their turn taken into the current.

Here then the formation of the first red corpuscles is certainly not endogenous.

The blastodermic cells destined to become red corpuscles ordinarily multiply to a greater or less extent by fission before undergoing hæmoglobic degeneration. The older the embryo grows, the more this segmentation appears to give place to cells of smaller diameter, and the more rapidly does this degeneration of the cells proceed.

At first the cells which go to form the hæmatids are very large, they pass into the circulating stream before the nucleus has disappeared, or at least before it has ceased to present the ordinary chemical characters of unclear substance, these cells become the large embryonic hæmatids. Later on segmentation goes on more rapidly, and the resulting cells are even smaller and the nucleus rapidly atrophies. This may go on in two ways, either the nucleus gradually diminishing in volume loses its chemical characters, takes on those of the protoplasm surrounding it (the final process in the hæmatids of oviparous animals, as I described above),

or else the nucleus undergoes a sort of breaking up, and its substance becomes dispersed in that of the protoplasmic body, so that it disappears altogether. This method, however singular it may appear, I have distinctly proved to be the case. Such is certainly the origin of the first definite hæmatids comparable to those of the adult which succeed the nucleated hæmatids.

As to the transformation, or rather the hæmoglobic degeneration of the protoplasmic body itself, it always goes on in the same way, the protoplasm becomes more and more homogeneous, more hyaline, more refractive. As long as the newly-formed hæmatid remains immotile its true form remains obscured by the contact and pressure of the neighbouring elements. The hæmatid is not really biconcave and discoidal either at the moment when it leaves or at the moment when it enters the circulation.

Very soon after the rabbit's embryo has attained a length of 22 mm. the vascular area ceases to be the seat of origin of the hæmatids. Lastly, it is interesting to note that during the whole of the first period of intra-uterine life globulets are not found in the blood, they only appear later on, and in abundance only in certain animals.

Hæmatogenesis in the Adult.—Where are hæmatids formed in the adult ? This is the great question which is occupying the attention at this moment of such a number of observers, and which has already been thought to have been solved over and over again.

Usually observers have tried to relegate this so-called hæmatopoietic function to certain organs or to certain tissues. I must now call special attention to these. This function in mammals has been successively attributed to lymphatic glands, to spleen, to the marrow of the bones, the lymphoid patches in the mesentery of the rabbit. There are yet the supra-renal capsules and thymus, do they also deserve to be studied from the same point of view ?

Lymphatic Glands.—No hæmatids are found in the lymph stream, the few which have been seen in fish are exceptional, and probably came into it accidentally. Lymph taken from the thoracic duct of a dog, with proper precautions, never contains hæmatids, although in operating carelessly on the horse the contents of the lymphatic vessel appear of a reddish colour ; this is due to the large quantity of hæmatids whose presence is caused by the careless operation. In the lymphatic glands the blood system is a closed system. Indeed, I have shown that it is not the same as the spaces constituting the lacunar tissue which makes a com-

munication between the afferent and efferent vessels.   If at
certain places these channels are definitely closed in, they
sometimes open directly into the follicular substance ; in short,
there is really no valid distinction to be drawn between the
two tissues which are described as forming the Malpighian
corpuscles, the lacunar tissue, and the follicular tissue.
Therefore the following may be taken as a plan of a Mal-
pighian corpuscle.   On the lymph paths proper (lacunar
tissue) are arranged certain cæcal prolongations (follicular
tissue), which are closed only at the periphery, but open on
the other hand into the paths where they are continuous
with them, by lacunæ, which are here large, but get narrower
and narrower towards the bottom of the cæca.

Beyond the point of insertion on the lymph path the
cæcum is definitely bounded, and bounds in its turn the
lymph path, or, in other words, the regions of lacunar tissue.
In these cæcal prolongations, as well as on the walls of the
trabeculæ of the so-called lacunar tissue, certain cells pro-
liferate, and masses of cells are seen developing in them
comparable to the nuclei of origin of the leucocytes, and
which are evidently destined to fall into the lymph current,
and form the latter.   But sometimes these same cells,
especially those in the lacunar tissue, undergo a different
development.   The protoplasmic body of the cell becomes
rounded, and presents, along with other granulations of un-
known nature, four, five, or six large granules, sometimes
almost polyhedral, of a substance having all the characters
of hæmoglobin.   These granules have been constantly taken
for hæmatids in the course of formation, or for hæmatids
collected together in cells to which amœboid movements
have been attributed, although no one has ever proved their
existence, forgetting that the ultimate origin of the hæma-
tids thus observed in contact with the cells in question has
yet to be explained.

The interpretation of this phenomenon seems simple
enough.   Hæmoglobin is not a special product of hæma-
tids ; it also occurs in the leucocytes of Semmer.   The large
granules of hæmoglobin found in the cells of the lymphatic
glands certainly do not originate elsewhere, and, moreover,
are not simply hæmatids absorbed by the cells of the no-
dules, just as they are not developing hæmatids.   The
presence of these granules of hæmoglobin always has the
effect, when they are abundant, of giving the tissue of the
Malpighian corpuscle a reddish tint, and even a decided red
colour, if the cells so altered are very numerous.   We
shall see presently to what errors this modification of the

tissue of the nodules, which has nothing whatever to do with hæmatogenesis, has given rise.

The Spleen.—The older observers, by experiments which I have repeated, proved beyond a doubt that the spleen was not essential in the regeneration of the blood after extensive hæmorrhage. I have already briefly described the structure of the tissue of the spleen. As I said, it is extremely probable that a large number of the hæmatids are normally arrested in it when they have lost their elasticity, which they do as they grow old, on account of the large amount of hæmoglobin then present. These old hæmatids, thus retained in the meshes of the splenic tissue, certainly help to give it its colour, which it possesses even when there is very little blood in it.[1]

The fact that the serum found in the splenic vein appears to be more yellow than that found in other vessels (G. E. Rindfleisch), might perhaps be explained by this breaking up of the old hæmatids, which are arrested in the parenchymatous tissue of the spleen.

Those who have attributed to the spleen a definite hæmatogenetic function, ought, at the same time, to remember that hæmatogenesis is no less active in mammals from which the spleen has been removed; they have gone further, and supposed that the spleen was helped by the mesenteric glands, and, indeed, even by the subperitoneal tissue! At least, it ought to be proved in this case that the glands and the cellular tissue have taken on the totally different histological structure of the spleen-parenchyma; and the stages of such a wonderful transformation, which would be one of the most curious known to anatomists, ought to be pointed out! On the other hand, it would be a senseless physiology which ascribed to two organs, of such essentially different structure and texture as these, the same function. It is really astonishing that biologists have accepted this singular idea of a vicarious action, as it has been called, on the part of certain organs which are supposed to take on, for the time being, the functions

[1] Probably the Malpighian corpuscles ought not to be looked upon as special structures, but simply as regions where the splenic tissue, more or less accidentally, has become impermeable to the blood which passes through the organ. In telcostian fishes these impermeable regions are not isolated, but form a thick net-work in the organ; moreover, in Amphibia there is a development of certain cells of the spleen, which recalls (at least, so far as one can judge without having specially studied it) the development of those cells, which I shall describe further on, in the marrow of the bones of mammals.

of other organs which have not the same anatomical structure.

The Marrow of the Bones.—Of all the questions which touch upon hæmatogenesis, perhaps the most delicate and, moreover, the most difficult to settle, is the part which is played by the marrow of the bones. Just now this hæmatopoietic function, which has been attributed to so many organs one after another, is ascribed to the marrow of the bones ; and it must be admitted there are more or less valid reasons for it. In the first place, in all mammals, without exception, the marrow of the bones preserves the characters which it has in the fœtus, that is to say, the marrow remains red, particularly in the bodies of the vertebræ.[1]

Even in mammals which have a large quantity of fat, *e. g.* Cetacea, I have ascertained that the marrow of the vertebræ and the spongy substance of the large bones of the limbs is red ; lastly, the red marrow cannot be removed from an animal in the way the spleen can, and the part it plays in hæmatogenesis experimentally judged.

In 1868 there was a dispute between MM. Neumann and Bizzozero, as to who had discovered in the red marrow of animals anatomical elements, the protoplasmic body of which presented the same characters as the substance of the hæmatids, but which at the same time possessed a nucleus. Stated thus, this fact, as pointed out by MM. Neumann and Bizzozero, is perfectly correct, but MM. Neumann and Bizzozero, each on their own side went further, and concluded that the red marrow was essentially hæmatopoietic, and that the elements that they pointed out were none other than young hæmatids in the course of development. This interpretation may be true, but it is not yet proved. To accept this hypothesis we must take it for granted that hæmatids are formed by the atrophy or hæmoglobic degeneration of the true cells of the marrow, and that they fall, after their nucleus has completely disappeared, into the blood-stream, just as the nuclei of origin fall (as I have shown above) into the lymph stream.

The first question that then suggests itself is—have the capillaries of the red marrow any walls? M. Hoyer, and more recently M. G. E. Rindfleisch, have declared that the medullary capillaries possess a wall. Moreover, M. Rustizky showed, in 1872, and I have since proved, that the medullary capillaries are certainly coated with endothelial cells.

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[1] Excepting in the last caudal vertebræ, where on the contrary it is extremely full of fat.

The first point we wished to know is then well established.

It is also certain that many of the marrow cells undergo a hæmoglobic degeneration altogether comparable to that undergone by the hæmatids in circulation in the blood of oviparous animals. The protoplasmic body, at first colourless and finely granular, soon becomes hyaline, coloured, and refractive. At the same time the nucleus gradually loses its chemical characters and finally disappears. The peculiar red colour of the marrow is due to the abundance of these elements.

The question so often discussed as to the essential identity of the marrow elements (medullo-celles of M. Ch. Robin) and the leucocytes is here beside the mark. Such identity is improbable, and in any case the " medullo-celles " never present the characteristic four nuclei found in the leucocytes of the lymphatics. We might here call to mind the words of the German author of one of the last works on the subject, " The term white corpuscle has become a sort of omnibus into which everything is thrust."

It is now well known that the medullary elements undergo hæmoglobic degeneration along with a disappearance of the nucleus.

In the less modified elements independent masses of hæmoglobin appear almost as large as hæmatids, but pushed out of shape by the neighbouring elements.

It has been proved that before this degree of degeneration is attained the masses of hæmoglobin contain a nucleus which disappears by gradual assimilation into the protoplasmic body as in the vascular area in Rodents, and not by going out of the cell, as has recently been stated (G. E. Rindfleisch). The question arises do these masses of hæmoglobin, which have been very properly called " medullary hæmatids," and which are moreover comparable in all respects to the hæmatids of birds, end their retrograde development by becoming dissolved, or do they fall into the blood stream ?

It is impossible to suppose that these elements can exhibit spontaneous movements, so as to admit of their displacing themselves, or becoming detached from the walls of the capillary, and finally passing through the endothelial cells, or pushing in between them by a sort of reversed diapedesis. One of the essential characters of hæmoglobic degeneration is the almost immediate loss of all power of amœboid movement in the protoplasm.

On the other hand, can we look to external forces to accomplish for the hæmatid this migration ? No ! for the

marrow is quite immovable and specially fixed in the solid substance of the bone.

Another question arises, if this degeneration were to go on at the same time in a number of elements bordering upon a capillary, the wall which, as we have seen, is formed of a single layer of endothelial cells, would break through sooner or later and allow the blood stream to take in these new hæmatids while they were yet undeveloped, while a new endothelial wall would have to form to cover up the space where they had been set free. In other words, would not the capillaries of bone be continually undergoing development or rather displacement in the medullary tissue? Nothing in such observations relating to this, as I for my part have been able to make, furnish any indication of anything of the kind taking place, or that the medullary cells while undergoing development come into any special relation with the capillaries.

Certain anatomists have thought that the marrow of the bones was modified after great hæmorrhage, and when the blood is being regenerated. All the experiments—which, however, have not been many—that I have made in this matter, have not shown this to be the case.

It must then be admitted, and I think we can go no further, that the marrow cells undergo *in situ* a hæmoglobic degeneration comparable to that taking place in the hæmatids of birds, and which also takes place in the marrow of the bones of birds before it has given place to the air cavities. In fact, marrow exists in the bones of reptiles and Amphibia, as well as in young birds, and yet no one has dreamt of ascribing any hæmatopoietic function to it. Fishes have no marrow in their bones.

To sum up:—the development of hæmatids among oviparous animals and that of the marrow cells are two processes which are in all points comparable to one another, just as the appearance of granules of hæmoglobin in the lymphatic gland is comparable to what happens in the leucocytes of Semmer.

It has been attempted to found an argument in favour of the hæmatopoietic function of the marrow on the existence, which has been proved from time to time, of cells in the blood, with a nucleus and a body containing hæmoglobin-cells consequently analogous to the hæmatids of birds, only often without a regular form.

It will be sufficient to draw attention to the extraordinary rarity of these elements which one hardly finds in one out of a hundred preparations. They are not moreover more

abundant in blood which is undergoing repair than in normal blood. Certainly it may be that they are marrow cells which have accidentally fallen into the blood current, but it is, perhaps, more logical to look upon them as leucocytes which have accidentally undergone a hæmoglobic degeneration after the fashion of marrow cells or the hæmatids of oviparous animals. In any case the extreme rarity of these elements takes away from them any value in the solution of the problem of hæmatogenesis.

Lymphoid Patches in the Rabbit.—MM. Ranvier and Hayem have recently held that hæmatids being devoid of nucleus were necessarily endogenous cellular productions.

M. Ranvier has brought to bear upon this subject his observations on the lymphoid patches in the mesentery of the rabbit where he has thought he has seen and has figured hæmatids originating in the midst of angioplastic cells destined to become the walls of the vessels. But admitting the perfect accuracy of the observations of such a skilful anatomist, it must be allowed it would be very difficult to ascribe to them such a character. In this case it would follow that the constant repair of the blood is necessarily bound up with the productions of new capillaries, and that the restoration of the blood after great hemorrhage is accompanied by a considerable extension of the capillary system ! It has not yet been shown that this is the case.[1]

Blood undergoing Repair.—It only remains to examine the conditions which blood presents among mammals while undergoing repair after great hæmorrhage. Observing the blood under these conditions one is struck with the extraordinary abundance of globulets, and above all, by the number of intermediate forms between the globulets properly so called and the elongated oval hæmatids which I described above ; and no doubt these are all the same, passing from one form to the other.

They are very easily seen in the dog. I have also observed them in the rat. It is only necessary to subject these animals to copious and frequent bleeding   The globulets of Donné (hæmatoblasts of M. Hayem) appear to me, as Zimmerman suspected, and as M. Hayem admitted, to be the true origin of the hæmatids of mammals. They represent the young state just as the microcytes of MM. Vanlair and Masius represent the old and decayed state.

[1] A thing which circumstances only very rarely allow one to study, and which would be of considerable interest in this connection, is the mode of repair of the uterine mucous coat during an intermenstrual period.

The globulet, which after its appearance in the serum is distinctly elongated, enlarges in all directions ; its substance, which at first perhaps appeared finely granular, becomes perfectly hyaline and refractive. It passes into the state of an oval elongated hæmatid, the long diameter of which is larger than the diameter of the discoidal hæmatids which have a slightly pronounced and thickened margin. The normal discoidal hæmatid thus represents a more advanced stage in the development of the cell, and is, in fact, its adult condition. To this succeeds a period of degeneration, during which the hæmatid becomes irregularly spherical and darker in colour, before disappearing altogether.

Origin of Globulets.—How do the globulets originate ? Are they, as M. Hayem wished to show, an endogenous product of certain cells which are at present unknown ? Or, on the contrary, must we recognise in these elements direct products, organic concretions, of a special kind originating in the blood plasma itself ? As a matter of fact their morphological characters, which are well known, do not allow of their being looked upon as amorphous deposits of albuminous matter. They have evidently a definite structure, and on this account are entitled to be called anatomical elements as well as the crystals of " otoconia " or the laminated fibres of connective tissue, if it is to be admitted that these latter form independently of the body of the cells which form the fibres.

The globulet exhibits the peculiarity common to it and to many cells of fixing or forming hæmoglobin. The gradual accumulation of the latter explains the growth of the corpuscle. The proportion and character of the hæmoglobin regulates the form of the corpuscle which at first is oval and afterwards discoidal. When there is a large amount of hæmoglobin present further growth stops. Thus the amount of hæmoglobin determines the limit of growth. This limit is, on the other hand, in direct relation with the minimum diameter of the vessels through which the hæmatid has to pass. I am well aware that if the evolution of the hæmatids is such as I have indicated, it goes against certain well-known facts of general anatomy. This, however, is not always a sufficient reason for rejecting a theory, which if it be true, would be, on the contrary, an explanation of what has been for a long time misunderstood. According to my hypothesis, the hæmatids of adult mammals would not be cells, would not even be derived from cells.

I may here remark that hæmoglobin should be regarded as a product of a cellular organism and not as an integral

part of the same. In this respect hæmoglobin behaves like fat, or like vitellin granules, or starch, &c. It might be even more exactly compared to chlorophyll, which sometimes appears in the midst of the cellular substance, and sometimes in solution in the body of the cell itself.[1]

Sometimes, in fact, the hæmoglobin appears stored up in the cell (leucocytes of Semmer, cells of lymphatic glands), and at other times diffused throughout the whole body of the cell, only in proportion as the substitution is more and more complete the cell body loses more and more its vital properties, properly so called ; it becomes inactive, and the nucleus disappears. Soon the whole is nothing but the wreck of a cell.

It appears, at the same time, that as more and more hæmoglobin is formed this living matter cannot continue to exist and disappears, and, as a consequence, the cell, which is reduced merely to the hæmoglobin which is deposited in it, is able to dissolve in the plasma, and so disappear.

All this tends to show that hæmoglobin is a very secondary product of the organism, and it would not be astonishing if this body, which results from extraordinary complex chemical actions going on in the blood plasma, were sometimes to form and be deposited in cells, properly so called.

As to the first appearance of the globulets in the midst of the blood plasma, it is not really more surprising than that of the fibres which appear in the same plasma when drawn from the vessels—fibres which, it must be remembered, have a definite morphological character.

To the globulet thus formed of albuminous substance (globulin) there is soon added a crystalline substance, or at least a substance having certain characters which belong to crystalline substances (hæmoglobin), and it is to this that the increase in volume is due. Such an increase of volume is, therefore, not a case of development in its technical sense, but simply a growth comparable to that of a group of bodies similarly formed by the union of albuminous compounds and cystalline compounds, which M. Harting and others have studied so carefully.

To sum up, it must be admitted that the origin of the hæmatids among adult mammals has not as yet been completely made out. Anatomists are divided between two

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[1] " It sometimes happens (among algæ) that the whole protoplasmic mass is coloured green excepting the most internal layer, the membranous layer, and certain isolated spots (many Zoosporæ, Palmellaceæ, gonidia of lichens." Sachs.

principal theories. Some, with MM. Neumann and Biz-zozero, distinctly attribute to the red marrow this function in the economy—the production of the hæmatids and the provision for the normal regeneration of the elements of the blood after accidental losses.

I think, on the contrary, with M. Hayem, that the hæmatids originate from the globulets of Donné ; only we differ as to the origin of these globulets.

Whilst M. Hayem looks upon them as the endogenous product of cells which are as yet unknown, it seems to me that everything goes to prove that they are formed in the blood plasma whilst in circulation, in a way which is more or less analogous to the formation of the filaments of fibrin in blood which has been drawn from the vessels.

On LIMNOCODIUM (*Craspedacustes*) SOWERBII, *a new* TRACHO-
MEDUSA *inhabiting* FRESH WATER.   By E. RAY LANKESTER,
M.A., F.R.S.   With Plates XXX and XXXI.

AT the meeting of the Royal Society on June 17th, and in
' Nature' of that date, I described in the following terms a re-
markable Medusa which I had received a few days before:

"On Thursday last, June 10th, Mr. Sowerby, the Secretary
of the Botanical Society of London, observed in the tank in the
water-lily house in Regent's Park some peculiar organisms, of
which he was kind enough to place a large number at my dis-
posal on the following Monday.

"The organism proves to be an adult Medusa, belonging to
the order Trachomedusæ, and the family Petasidæ, of Haeckel's
system.   It comes nearest among described genera to Fritz
Müller's imperfectly known Aglauropsis, from the coast of
Brazil.

"The most obviously interesting point about the form under
notice is that it occurs in great abundance in perfectly fresh
water, at a temperature of 90° F.   Hitherto no Medusa of any
order has been detected in fresh water.[1]

"It is exceedingly difficult to trace the introduction of this
animal into the tank in the Regent's Park, since no plants have
been recently added to the lily-house, and the water is run off
every year.   Probably a few specimens were last year, or the
year before, present in the tank, and have only this year multi-
plied in sufficient abundance to attract attention.   Clearly the
Medusa is a tropical species, since it flourishes in water of this
high temperature (90° F.)

"Mr. Sowerby has observed the Medusa feeding on Daphnia,
which abounds in the water with it.

"The present form will have to be placed in a new genus, for
which I propose the name Craspedacustes.[2]

"It presents the common characters of the Trachomedusæ (as
distinguished from the Narcomedusæ) in having its genital sacs
or gonads placed in the course of the radial canals.   It agrees
with both Narcomedusæ and Trachomedusæ in having endoder-
mal otocysts—and further presents the solid tentacles with car-
tilaginoid axes, the centripetal travelling of the tentacles, the
tentacle-rivets (Mantelspangen), the thickened marginal ring to
the disc (Nesselring) observed in many Trachomedusæ.

[1] Except, perhaps, some estuarine forms (Crambessa and others).
[2] Κρασπεδακουστης, one who hears by means of the κρασπεδον, or
velum.

" Amongst Trachomedusæ, Craspedacustes finds its place in the Petasidæ, which are characterised as ' Trachomedusæ with four radial canals, in the course of which the four gonads lie ; with a long tubular stomach and no stomach stalk.'

" Amongst Petasidæ it is remarkable for the great number of its tentacles, which are *all* solid, and for its very numerous otocysts.   Further it is remarkable amongst all Hydromedusæ (velate Medusæ exclusive of Charybdæa) for the fact that centrifugal radiating canals pass from the otolithic concretions which they enclose *into the velum where they end cæcally*.

" The genus may be characterised as follows :

" MOUTH quadrifid, with four per-radial lobes.

" STOMACH long and tubular, projecting below the disc.

" DISC, flattened.

" RADIAL CANALS, four terminating in the marginal canal.

" MARGINAL (Ring) CANAL, voluminous.

" CENTRIPETAL CANALS (such as those of Olindias, Geryonia, &c.) absent.

" TENTACLES, solid ; in three sets, the horizons of the insertion of which are superimposed : —

" (1.) A highest (nearest the umbrella-pole) set of *four* large per-radial tentacles—primary tentacles.

" (2.) A second tier of *twenty-eight* or more medium-sized tentacles placed between these in four groups of seven—secondary tentacles.

" (3.) A third tier of *one hundred and ninety-two* or more small tentacles placed in groups of six between the last—tertiary tentacles.

" TENTACLE-RIVETS (Mantelspangen) connecting the roots of the tentacles with the marginal ring (Nesselring) are connected with all the tentacles.

" OTOCYSTS placed along the line of insertion of the velum : about eighty in number, from sixteen to twenty between each pair of perradial tentacles, arranged in groups of two or three between the successive secondary tentacles.

" VELAR CENTRIFUGAL CANALS are present, passing from the otocysts—one from each otocyst—into the velum, and there ending blindly.   They appear to correspond in character to the *centripetal* canals found in other Trachomedusæ in the disc. Their presence constitutes the chief peculiarity of the genus Craspedacustes, and may necessitate the formation of a distinct family or suborder for its reception.

" (The minute characters of the otocysts, and their relation to these canals, is now in course of investigation.)

" OCELLI are wanting.

" GONADS. Four oval sacs depending into the cavity of the subumbrella from the four radial canals.

" The above characters are derived from the examination of adult male specimens, which were freely discharging ripe, actively motile spermatozoa.

" *Species.—C. Sowerbii.*

" I name the species in honour of Mr. Sowerby, who discovered it, and to whose observation and courtesy zoologists are much indebted. The sole character, which I can give as specific over and above the generic characters summarised above, is that of size. The diameter of the disc does not exceed half an inch.

" *Locality*.—The water-lily tank in the gardens of the Botanical Society, Regent's Park, London. Very abundant during June, 1880. Probably introduced from the West Indies."

On June 17th, the same day as that on which the above appeared in ' Nature,' the same organism was described by Prof. Allman to the Linnæan Society ; and in the following week (June 24th) an article was published by Prof. Allman in ' Nature,' which was an extension of the paper read by him to the Linnæan Society, including " some facts, in addition to those contained in his original paper." Professor Allman's observations agree very closely with my own reproduced above, though we differ on one or two important points, and especially on the relationship of the new Medusa to Trachomedusæ, on the one hand, and Leptomedusæ on the other, with which latter group Professor Allman thinks it has most affinity.

Inasmuch as Professor Allman gave his account of the new Medusa at the Linnæan Society on the same day as that on which mine was published in ' Nature,' and on account of the regard which all zoologists must feel for him, I propose as a mark of respect to him to accept the generic name Limnocodium which he bestowed upon the new Medusa in place of Craspedacustes which has the right of priority. At the same time I shall maintain the specific title ' Sowerbii,' in recognition of Mr. Sowerby's discovery and his valuable observations on the habits and on the embryonic condition of this Medusa.

In the following pages I propose to describe a few important facts relative to the structure of Limnocodium, which in my judgment suffice to establish its position as one of the Trachomedusæ, though it is quite a peculiar form and very possibly is either the isolated representative of an archaic type of that order, or has degenerated in connection with its exceptional life-conditions, namely, those of fresh water.

The facts to which I shall on the present occasion draw attention, relate to the structure of the marginal ring of the disc, the insertion of the tentacles and the structure and development of

the marginal bodies, and their capsules (referred to above as " otocysts " and " velar canals "). I am also able to give some conclusive observations on the embryonic condition of Limnocodium. I have not, as I have the intention of doing later, studied this Medusa by means of sections, but in the living state and with the aid of that invaluable reagent, osmic acid.

The account of the structure of this Medusa, published by Professor Allman, is as follows :

" The umbrella varies much in form with its state of contraction, passing from a somewhat conical shape with depressed summit through figures more or less hemispherical to that of a shallow cup or even of a nearly flat disc. Its outer surface is covered by an epithelium composed of flattened hexagonal cells with distinct and brilliant nucleus. The manubrium is large ; it commences with a quadrate base, and when extended projects beyond the margin of the umbrella. The mouth is destitute of tentacles, but is divided into four lips, which are everted and plicated. The endoderm of the manubrium is thrown into four strongly-marked longitudinal plicated ridges.

" The radial canals are four in number; they originate each in an angle of the quadrate base of the manubrium, and open distally into a wide circular canal. Each radial canal is accompanied by longitudinal muscular fibres, which spread out on each side at the junction of the radial with the circular canal.

" The velum is of moderate width, and the extreme margin of the umbrella is thickened and festooned, and loaded with brownish-yellow pigment cells.

" The attachment of the tentacles is peculiar. Instead of being free continuations of the umbrella margin, they are given off from the outer surface of the umbrella at points a little above the margin. From each of these points, however, a ridge may be traced centrifugally as far as the thickened umbrella margin ; this is caused by the proximate portion of the tentacle being here adnate to the outer surface of the umbrella. It holds exactly the position of the " mantelspangen " or *peronia*, so well developed in the whole of the Narcomedusæ of Haeckel, and occurring also in some genera of his Trachomedusæ. Its structure, however, differs from that of the true *peronia*, which are merely lines of thread-cells marking the path travelled over by the tentacle as the insertion of this moved in the course of metamorphosis from the margin of the umbrella to a point at some distance above it, while in Limnocodium the ridges are direct continuations of the tentacles whose structure they retain. They become narrower as they approach the margin.

" The number of the tentacles is very large in adult specimens. The four tentacles which correspond to the directions of

the four radial canals or the perradial tentacles are the longest and thickest. The quadrant which intervenes between every two of these carries, at nearly the same height above the margin, about thirteen shorter and thinner tentacles, while between every two of these three to five much smaller tentacles are given off from points nearer to the margin, and at two or three levels, but without any absolute regularity; indeed, in the older examples all regularity, except in the primary or perradial tentacles, seems lost, and the law of their sequence ceases to be apparent.

"I could find no indication of a cavity in the tentacles : but they do not present the peculiar cylindrical chorda-like endodermal axis formed by a series of large, clear, thick-walled cells which is so characteristic of the solid tentacles in the Trachomedusæ and Narcomedusæ. From the solid tentacles of these orders they differ also in their great extensibility, the four perradial tentacles admitting of extension in the form of long, greatly attenuated filaments to many times the height of the vertical axis of the umbrella, even when this height is at its maximum ; and being again capable of assuming by contraction the form of short thick clubs. Indeed, instead of presenting the comparatively rigid and imperfectly contractile character which prevails among the Trachomedusæ and the Narcomedusæ, they possess as great a power of extension and contraction as may be found in the tentacles of many Leptomedusæ (Thaumantidæ, &c.). These four perradiate tentacles contract independently of the others, and seem to form a different system. All the tentacles are armed along their length with minute thread-cells, which are set in close, somewhat spirally-arranged warts.

"The lithocysts or marginal vesicles are, in adult specimens, about 128 in number. They are situated near the umbrellar margin of the velum, between the bases of the tentacles, and are grouped somewhat irregularly, so that their number has no close relation with that of the tentacles. They consist of a highly refringent spherical body, on which may be usually seen one or more small nucleus-like corpuscles, the whole surrounded by a delicate transparent and structureless capsule. This capsule is very remarkable, for instead of presenting the usual spherical form, it is of an elongated pyriform shape. In its larger end is lodged the spherical refringent body, and thence becomes attenuated, forming a long tubular tail-like extension, which is continued into the velum, in which it runs transversely towards its free margin, and there, after usually becoming more or less convoluted, terminates in a blind extremity.

"The marginal nerve-ring can be traced running round the

whole margin of the umbrella, and in close relation with the otolitic cells. Ocelli are not present.

"The generative sacs are borne on the radiating canals, into which they open at a short distance beyond the exit of these from the base of the manubrium. They are of an oval form, and from their point of attachment to the radial canal hang down free into the cavity of the umbrella. Some of the specimens examined contained nearly mature ova, which, under compression, were forced from the sac through the radial canal into the cavity of the stomach.

"While some of the characters described above point to an affinity with both the Trachomedusæ and Narcomedusæ, this affinity ceases to show itself in the very important morphological element afforded by the marginal bodies. In both Trachomedusæ and Narcomedusæ the marginal bodies belong to the tentacular system ; they are metamorphosed tentacles, and their otolite cells are endodermal, while in the Leptomedusæ, the only other order of craspedotal Medusæ in which marginal vesicles occur, these bodies are genetically derived from the velum. Now, in Limnocodium the marginal vesicles seem to be as truly velar as in the Leptomedusæ. They occur on the lower or abumbral side of the velum, close to its insertion into the umbrella, and the tubular extension of their capsule runs along this side to the free margin of the velum, while the delicate epithelium of the abumbral side passes over them as in the Leptomedusæ. It is true that this point cannot be regarded as settled until an opportunity of tracing the development is afforded ; but in very young specimens which I examined I found nothing opposed to the view that the marginal vesicles were derived, like those of the Leptomedusæ, from the velum.

"If this be the case Limnocodium will hold a position intermediate between the Leptomedusæ and the Trachomedusæ; but as the greatest systematic importance must be attached to the structure and origin of the marginal vesicles, its affinity with the Leptomedusæ must be regarded as the closer of the two."

It will be observed that, in opposition to the results which I had published in the previous week, Professor Allman holds that the structures which I had identified with the "peronia" (tentacle-rivets or Mantelspangen) of Trachyline Medusæ are not "true peronia ; " further, that the tentacles, though solid, do not present "the chorda-like endodermal axis" characteristic of Trachomedusæ and Narcomedusæ, and lastly, that the otolite cells are not endodermal, as in Trachyline Medusæ, but are derived from the ectoderm of the velum, as in Leptomedusæ, and on this ground especially he holds that the affinity of the new

form must be regarded as closer with the Leptomedusæ than with Trachomedusæ.
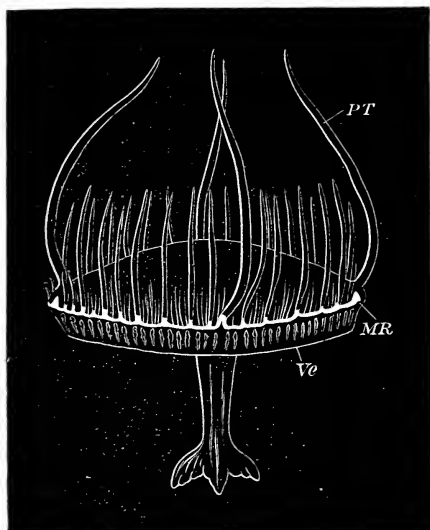
FIG. 1.—*Limnocodium Sowerbii*, as seen floating. Magnified five diameters. *MR,* marginal ring; *Ve,* velum, with tubular otocysts; *PT,* perradial tentacle.

In the present communication I shall apply myself to these
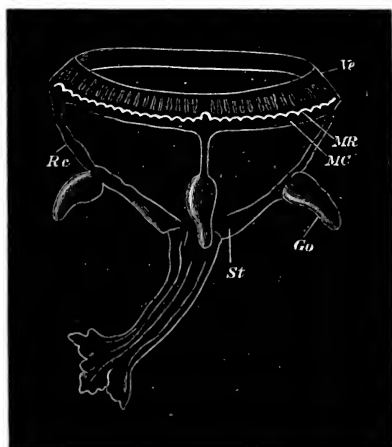
FIG. 2.—*Limnocodium Sowerbii*, with retroverted disc. *MR,* marginal ring; *MC,* marginal canal; *Rc,* radial canal; *St,* stomach; *Go,* gonad; *Ve,* velum. Magnified five diameters.

points, and first of all discuss the structure of the peronia and tentacles.

Naked-eye appearances.—When the Medusa is seen swimming in the water, the observer is at once struck by the manner in which the tentacles are carried. Though they can be depressed and directed towards the subumbrella surface, they are, as a rule, carried nearly at right angles to the horizontal plane of the disc and directed upwards. This carriage of the tentacles is more particularly noticeable when the Medusa is at rest after a certain number of pulsations, but is also seen between each one of a series of pulsations. An opaque crenated line along the margin of the disc (fig. 1, *MR*.) is very readily observed, even without any magnifying glass. This line is the marginal ring, consisting externally of ectoderm cells charged with thread-cells, and more deeply strengthened and supported by the thickened wall of the marginal or ring canal, the endoderm cells of which are converted into a cartilaginoid tissue, having a yellowish-green tint when seen by transmitted light with the microscope. These yellowish cartilaginoid cells have led Professor Allman to speak of the margin of the disc as being " loaded with brownish-yellow pigment-cells."

The layer of thread-cells along the margin of the disc, although not bulky in this small form, yet undoubtedly corresponds to the " nettle-ring " (Nesselring) of the typical Trachyline Medusæ, whilst the subjacent cartilaginoid tissue corresponds to the "ring-cartilage " (Ringknorpel) of the same group.

The crenations or centripetal projections of the opaque marginal ring are in relation to the insertions of the tentacles. They have the same superficial structure as the superficies of the ring itself, and, so far as any definition can be given to the term " peronium," the centripetal prolongations of the nettle-ring passing from that structure to the point where a tentacle springs from the disc, are entitled to the name. Below the peronia we find in each case an extension of the peculiarly modified endodermal cartilaginoid tissue of the ring-canal, which passes with rapid gradation into the cartilaginoid tissue of the tentacle-root.

The power of contraction and expansion possessed by the tentacles is no doubt, as Professor Allman has pointed out, greater than that possessed by the most highly differentiated of the Trachyline Medusæ. At the same time the stiff erect carriage to which I have alluded, is quite unlike anything seen in Leptomedusæ, and as I shall point out below the solid axis of the tentacles, though not so firm a tissue as the corresponding tissue of Cunina, is very appropriately described by the term " cartilaginoid," and more distinctly resembles " notochordal tissue "

than does the single row of thickened cells which is found in the
larger and typical Trachyline Medusæ.

The woodcut (fig. 2) represents the Medusa in an attitude
which it not unfrequently assumes when at the bottom of the
vessel in which it is confined. When this retroversion of the
disc is effected, the velum may take the position of a band ex-
ternal to the margin of the retroverted disc, or may also be
retroverted, as in the figure. In this case the tentacles are all
more or less concealed in the concavity of the retroverted disc,
and accordingly I have not represented them in the figure at all.
When in this position the Medusa supports itself on the manu-
brium as a stalk, exerting a feeble adhesive action by means of
the quadrifid lobes of the oral aperture.

This figure also serves to show the position and form of the
gonads (*Go*) on the radiating canals, which are omitted in
figure 1.
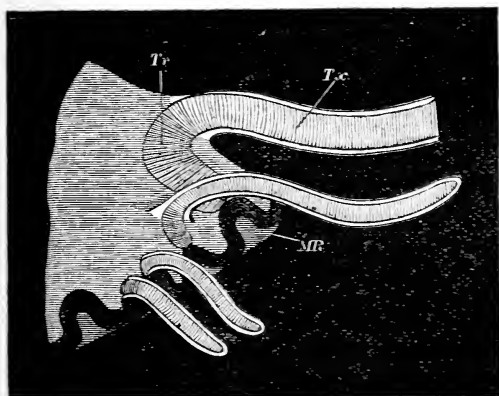
Fig. 3.—Diagram of margin of the disc to show tentacles and marginal
ring. *MR*, marginal ring; *Tx*, tentacle-axis; *Tr*, tentacle-root.

Insertion of the tentacles.—The tentacles do not arise from
the edge of the disc, but in three more or less distinct tiers,
indicating a *relative* centripetal movement of the tentacles
during the growth of the disc, as in many Trachyline Medusæ.

The four perradial tentacles are furthest from the marginal
ring and their free portion rises from the jelly-like convex surface
of the disc at some distance from the margin. At the same time
the cartilaginoid axis of the tentacles is continued beneath the
ectodermal tissues centrifugally towards the cartilaginoid sub-
stance of the marginal ring. It is this structural arrangement
which favours the peculiar vertical carriage of the tentacles,
since if they are to be directed downwards towards the mouth

a considerable flexure of the cartilaginoid axis at the point
where its free and inserted portions unite, becomes necessary.

This is shown in the diagram woodcut, figure 3, where tenta-
cles of three horizons are shown bent mouthwards by the
pressure of the cover-glass. The recurved axis-roots (*tr*) of the
two larger tentacles are seen by transparency through the
substance of the disc passing towards the margin, where they
become continuous with the centripetal upgrowths of the tissue
of the marginal ring (*MR*).

The smaller tentacles spring almost directly from the mar-
ginal ring, and have therefore no length of axial root. This
enables the small (which are the younger) tentacles to assume
more readily the dependent mouthwardly directed carriage ;
they are not mechanically directed into the vertical upright
position by the elasticity of a vertical axial root.

*Tissue of the nettle-ring and peronia, and of the cartilage-
ring and tentacle-roots.*—In order to obtain a satisfactory view
of these structures with the microscope, it is necessary to get
the margin of the disc in such a position when prepared for
examination that the optical plane is tangential to the concave
surface of the disc. Under these circumstances such a view as
that drawn in Pl. XXX, fig. 4, may be obtained. This re-
presents the part as seen from the supraumbrellar surface. In
Pl. XXX, fig. 5, I have represented the same parts seen with
a deeper focussing, the optical plane now passing near to the
subumbrella surface.

The surface of the marginal ring and peronia is seen to be
formed by a continuation of the large-celled ectodermal tissue
of the supraumbrellar surface, but the cells which constitute
it differ from those of the general surface in being loaded with
thread-cells. The continuous marginal ring of nettle-cells thus
constituted is entitled to the name of "nettle-ring," though it is
only one cell deep and does not exhibit the marked specialisation
and thickening observed in some Trachomedusæ and Narcome-
dusæ. It may be regarded as a rudimentary condition of the
structure which is more highly developed in other Trachyline
Medusæ.

The same simple but, as it appears to me, distinct character,
is exhibited by the prolongation of this nettle-ring centripetally
towards the point where the tentacle becomes free of the disc.
The masses of thread-cells forming these little " peronia" are not
merely due to the continuation of the character of the ectoderm
cells which clothe the free portion of the tentacle. For the first
portion of the free part of the tentacle is almost devoid of thread
cells.

It is not a little difficult, on a first examination of this region,

to separate the superficial ectodermal cells with their refringent nematocysts (thread-cells) from the immediately subjacent tissue, the cells of which have a yellowish-green tint, and are also somewhat refringent.

By careful focussing this difficulty is overcome, and it is then found that beneath the nettle-ring is a peculiar tissue, which forms the abumbral wall of the marginal canal (Pl. XXX, fig. 5).

The marginal canal itself is wide, and capable of distension on its abumbral side, where the wall is formed by cells of a very different character from that of those which underlie the nettle-ring. In fig. 5 the superior wall of the marginal canal (MCa) is seen in section, showing the peculiar soft clear (ciliated) cells containing yellowish-green granules, by which it is here lined internally. The modified cartilaginoid cells of the abumbral wall are limited *inferiorly* (*i. e.* taking the mouth as indicating the inferior surface of the organism) by a strongly-marked row of quadrate cells, which form a slight ridge, and are especially observable by their complete and uniform pigmentation (Pl. XXX, fig. 3 VMC, fig. 4 VMC, fig. 5 MCp)—yellowish green or greenish brown. These cells form a dark line, which can always be recognised along the insertion of the velum, and accordingly I distinguish them as velo-marginal endoderm.

The relations of these parts are exhibited, in a diagrammatic form, in Pl. XXX, fig. 3. A comparison of this figure, with the surface view and the optical section, will, I hope, serve to make the structure comprehensible.

The true character of the tissue formed by the endodermal cells of the abumbral wall of the marginal canal is best appreciated by tracing the gradual transition of the tentacle root into this tissue.

The axis of the tentacle is formed by entoplastic cartilage cells, that is to say, cells the protoplasm of which undergoes a cartilaginoid metamorphosis *within* the cell area, as in the notochord of Vertebrata.

The tissue very closely resembles that of the notochord of a young Lamprey. It differs in the smaller density of the cartilaginous deposit, as also in the greater abundance of the cells, from that of typical Trachyline Medusæ, and resembles that of the solid tentacles of many hydroid polyps. At the same time there can be no question that these tentacles more closely resemble those of Trachomedusæ than they do those of Leptomedusæ, since in all Leptomedusæ the tentacles are hollow, whilst in Trachomedusæ the tentacles are very usually solid, as they are here.

In that part of the tentacle axis which is plunged in the disc,

and which is called "tentacle root," we find the same large cells
as those in the axis of the free part.

The root at first expands a little at its insertion into the disc,
but tapers as it approaches the wall of the marginal canal, and
becomes rounded off (see Pl. XXX, fig. 5 *TR*, and woodcut,
fig. 3). There is, however, no discontinuity of the tissue where
the tentacle root ceases and the marginal ring commences, but
rather a very rapid transition or change in the form of the en-
dodermal cells, which equally constitute the tentacle axis and
the marginal cartilage ring.

The cells, in fact, diminish very much in size whilst exhibit-
ing a characteristic polygonal form (Pl. XXX, fig. 6). In
place of the large vacuoles filled with gelatinous substance which
are presented by the large cells of the tentacle-axis, we find,
within the small polygonal cells, block-like deposits, one or two
to a cell, of a pale greenish tint and homogeneous structure.
The greenish colour of the contents of these cells shows through
the superficial ectoderm cells, and gives the whole "marginal
ring" a greenish-yellow appearance. It is most strongly de-
veloped in the velo-marginal cells which form an inferior limit
to these modified endodermal cells of the ring-canal's wall.

It cannot be disputed that in the character of these cells, as
compared with those of the adumbral wall of the marginal canal
(Pl. XXX, fig. 7), we have a very well-marked distinction.
These adumbral cells are metamorphosed by the intracellular
deposit of a homogeneous greenish substance, so as to form a
sort of cartilaginous tissue, which acts the part of and morpho-
logically represents the "cartilage-ring" of higher Trachyline
Medusæ. In order to appreciate their special character[1] it is
only necessary to compare them with the lining cells of the
adumbral wall of the marginal canal (Pl. XXX, fig. 7).

I am not able at present to state definitely whether the simple
rudimentary cartilage-ring of our Medusa is the *actual* limiting
wall of the ring-canal, or whether a second layer of endoderm
cells exists deeper than it, and actually bounding the cavity of
the ring-canal.

This question I hope to decide by the preparation of sections.

*Comparison of the arrangement of the tissues of the marginal
ring and tentacle root with those of typical Trachyline Medusæ,
and with structures found in the Leptomedusæ.*—If we compare
with what has just been described the arrangement of the homo-
logous parts in such a Trachyline Medusæ as Cunina, we find
very considerable differences, which, however, appear to me to be
differences of degree, our new Medusa exhibiting in a simple form

[1] Cells of the same character as those of the endoderm of the marginal
ring occur in parts of the stomach-wall.

conditions, which are highly specialised and developed in such forms as Cunina.

In the first place the tentacle-roots of Cunina, instead of passing centrifugally towards the margin of the disc, have rather a centripetal direction.   This is simply due to the fact that the marginal ring and its tissues are so largely developed as to push the tentacle root towards the centre of the disc; and is not a difference of a fundamental character.

Further, the cartilage of the ring is much more largely developed in Cunina than in the new Medusa, and acquires the importance of a well-marked skeletal ring, which it is only (as it were) beginning to assume in Limnocodium.   The same spe-cialisation of the nettle-ring and of the peronia is observable in Cunina as compared with Limnocodium, but does not invalidate the claim of the simpler structures found in the latter to recognition.

It appears exceedingly probable that the Trachyline Medusæ are the modified descendants of such simpler forms as the Leptomedusæ, and if we search amongst the latter for conditions approaching those exhibited by the tentacle-roots and marginal ring so frequent among the former, the nearest case which we can find is that of the genus Laodice, one of the Thaumantidæ of Haeckel's system.   Here the tentacles do not spring from the margin of the disc freely, but from the convex surface of the disc, the axis of the tentacle being prolonged as a root to join the wall of the marginal canal, which has a cartilaginoid character.   Nevertheless, in Laodice the tentacles are hollow, and there does not appear to be any structure corresponding to peronia.

From the consideration of the characters of the tentacles and marginal ring alone, I think that we should be led to the conclusion that Limnocodium is a representative of the Trachomedusæ in an early or archaic phrase of differentiation, already distinctively Trachomedusan, but not far advanced on that path.

We have next to see what indications a closer examination of the marginal bodies (otocysts) may give us, and we shall find that they confirm very distinctly the conclusion already enunciated.

Development of the marginal bodies.—The marginal bodies of Limnocodium vary in number, as do the tentacles, according to the size of the specimens examined.   I have seen as few as fifty in specimens measuring one third of an inch across the disc, and as many as 120 in larger specimens.   Even in large specimens the marginal bodies are continually being developed, so that there is no difficulty in obtaining that "opportunity for tracing the development" which Professor Allman has stated to be desirable.

The marginal bodies consist of a spherical refringent body

averaging $\frac{2}{10}$,$\frac{2}{000}$ths of an inch in diameter, enclosed in an elongated tubular capsule. The refringent body is situated at the line of junction of the velum with the marginal ring, within the slightly enlarged termination of the tubular capsule.

The tubular capsule is continued thence radially to the extreme margin of the velum, whence I have termed these capsules " velar canals" (Pl. XXX, fig. 1).

The refringent body belongs essentially to the abumbral side of the velum—that which is adjacent to the tentacles—as will be described below, whilst the capsule is entirely formed by the ectodermal cells of the abumbral surface of the velum.

The refringent bodies and their capsules form interruptions in a cellular band of thickened ectodermal tissue, which runs along the line of junction of the velum and disc peripherally to the strongly-coloured velo-marginal cells of the endoderm of the marginal ring.

The appearance of this ring of transparent, colourless cells, and the relation of the marginal bodies and their capsules to it, is seen in Pl. XXX, fig. 5, x. The transparent ring (x) is undoubtedly the representative of the abumbral nerve-ring of other Hydromedusæ; but I have not yet made that special histological study of its elements which would enable me to say how much of the ring is to be regarded as nervous tissue, and how much is unspecialised ectoderm.

The tubular capsules of the marginal bodies are often to be found in an incomplete state of development, and it is then possible to observe the process by which they increase in length, so as to reach finally the edge of the velum. The tubular capsules occupy a position between the abumbral and adumbral ectoderm layers which constitute the velum, and they actually stand out on the abumbral surface of the velum as ridges raising up the abumbral ectoderm of the velum, and separating it entirely from the adumbral ectoderm in the form of a delicate membrane. This is well seen when the velum is folded on itself, as not unfrequently happens when a specimen of the Medusa is placed beneath a cover glass, as shown in Pl. XXX, fig. 2.

If the tubular capsules be now carefully studied it is not difficult to make out that they increase in length, *not* by the simple growth of the already existing capsule, but by *the fusion with the capsule of vacuolated cells belonging to the abumbral layer of the velum* (fig. 2 *vac*).

The vacuolated cells of the abumbral ectoderm layer of the velum may be seen in various regions of the velum, ready, as it were, to fuse with the capsules, should their growth favour such an occurrence. The tendency to fuse on the part of the cap-

sules is indicated in a rare phenomenon, observed by my assistant (Mr. Bourne), namely, the fusion of neighbouring tubular capsules, an instance of which is shown in Pl. XXX, fig. 1.

Deformities and abnormalities of many kinds are not unfrequent in the capsules, the marginal bodies themselves, and the tentacles. The vacuolated cells of the abumbral velar surface appear to stand out from the stratum in which they originally lay, and sometimes to form separate vesiculi, which may or may not fuse with the growing capsule of the refringent body.

The adumbral ectoderm is complete and continuous *beneath* the tubular capsules, its circular muscular fibrils being traceable in every part beneath (that is, on the adumbral surface of) the tubular capsules (see Pl. XXX, fig. 2 *musc*).

The development of the velar canals or tubular capsules of the marginal bodies is, then, shown to be due to the fusion of vacuolated cells of the abumbral ectoderm of the velum.

It remains to be shown what is the first origin of the capsule and of the refringent body itself.

In Pl. XXXI, figs. 10 to 20, I have represented a number of stages in the development of the refringent body, and it is perhaps as well at once to say that the refringent body is nothing more nor less than a modified solid tentacle. It consists in the fully formed state of a number of cortical cells enclosing four, six, or eight large refringent cells (Pl. XXXI, figs. 10, 11). The cortical cells correspond to the ectoderm of a tentacle, and the medullary refringent cells to the endodermal axis of a tentacle, the whole group of cells being tightly pressed together so as to form a spherical refringent body. The cortical as well as the medullary cells of the free hemisphere of the body are highly refringent.

There is not in the refringent body of Limnocodium *any separate concretion, any otolith in the strict sense of the term.* The whole structure is purely cellular, and consists entirely of nucleated cells which can be isolated and recognised as distinct cell elements by means of reagents.

In this respect the refringent body of the new Medusa appears to differ from any previously described organ of the kind. Certainly it differs entirely from any marginal body either of Leptomedusæ or of Trachyline Medusæ described by the Hertwigs in their recent work 'Nervensystem der Medusen.' There seems very small justification for regarding the refringent body of Limnocodium as an auditory organ at all. It consists simply and solely of nucleated cells, the substance of which is of a highly refringent nature. "Refringent bulbs" would be a sufficiently appropriate name for these bodies. The cells which correspond to the tentacle ectoderm, and which I term "cortical

cells," are very delicate, often much stretched and translucent, but in order, as it were, to fill up the interstices of the spherical mass, they are sometimes enlarged in places and refringent like the medullary cells.

The cortical cells are, however, never so distinctively modified as the medullary cells. There is a peculiar brownish colour and a refringent appearance about the medullary cells, especially after treatment with osmic acid, which is not to be noticed in the cortical cells.

The medullary cells sometimes contain one or two greenish-yellow granules (figs. 8, and 15 *gr*), which are identical in appearance with the granules found in the endoderm cells of the marginal ring canal *from which they are derived*.

By searching along the velo-marginal line it is easy in some specimens to find refringent bodies and their capsules in all stages of development. The earliest indication which I have obtained of this development is a protrusion of the endodermic cells of the marginal canal into the ring or band (*x*) of colourless cells, which is ectodermal in origin, and probably to a large extent nervous in character. This protrusion (Pl. XXXI, figs. 12, 13), lifts the ectodermal tissue in front of it, and *one* of the in-growing endodermal cells enlarges and becomes highly charged with fine granules, and sometimes with coarser, green-coloured granules also. As far as I have been able to ascertain, this enlarged endodermal cell, which frequently is coloured like the other endo-dermal cells of the ring-canal, is the mother-cell, from which the axial or medullary cells of the refringent body are developed (see Pl. XXX, figs. 14, 17, 18, *axen*). The ectodermal tissue into which this enlarged cell has protruded now grows around it in a very re-markable way. In some cases it seems to form at first a complete investment of small cells—the cortical layer—enclosing the en-dodermal medullary cell. But this appears from such a con-dition as that represented in Pl. XXXI, fig. 14, not to be the actual mode of growth. The ectodermic layer, pushed forward by the axial outgrowth of endoderm, arranges itself in such a way as to form a cortical layer to the medullary cell, and at the same time a capsule embracing the central body formed by these two sets of cells (see Pl. XXXI, figs. 15, 16). From henceforth the medullary cell has merely to divide and give rise to the four or eight highly refringent medullary cells of the fully-formed re-fringent body, and the capsule has only to expand and increase its area by the agglutination of vacuolated ectoderm cells in the manner described above.

The ectodermic cortical investment of the highly refringent central or medullary cells of the marginal body is complete; the endodermic medullary cells are entirely cut off from their

seat of origin ; but the cortical cells rest upon the tissue of the transparent marginal ring x, and have some kind of attachment to that ring, though I am not able to point out any definite nerve-fibres. That the attachment is very slight is shown by the fact that sometimes a marginal body (refringent body) becomes broken off from its seat of origin, and falls into the tubular capsule. I have met with such a detached refringent body enclosed in a constriction of the peripheral extremity of a capsular tube (Pl. XXXI, fig. 20). This detached refringent body was remarkable for the fact that it had undergone an abnormal development, and was actually a *cyst* with an internal cavity, into which projected the large refringent cells, whilst it was clothed externally by the cortical cells.

That the refringent bodies in themselves, irrespective of the tubular capsules, correspond morphologically to solid tentacles is proved by an occasional abnormality. In place of a normal refringent body I have found rarely a tentacle-like body one of which is drawn in Pl. XXXI, fig. 18. This small tentacle was in exactly the same position as a refringent body. It is seen to have in its axis one large and apparently two smaller granular cells similar to those which are protruded from the endoderm of the ring-canal to form the medullary cells of the normal refringent body. At the same time the ectoderm which clothes these axial cells is free and superficial. Instead of forming a *capsule,* the ectoderm cells have simply invested the axial growth as they do in the case of a tentacle.

Further, I have observed another abnormality of an exactly complementary character, namely, the commencement of the tubular ectodermal capsule without any refringent body to occupy its proximal extremity (Pl. XXXI, fig. 19). The existence of these two elements separately one from the other, justifies us in regarding them as two distinct structures which have united to form the complete " refringent bulb and sac " of our new Medusa.

Have we anything parallel in Leptomedusæ or in Trachomedusæ for the two distinct elements, the modified tentacle and the velar ectodermal capsule, above described ? Assuredly there is no parallel at present described among Leptomedusæ. Among Trachomedusæ, however, we find a very exact parallel. Although in no other Trachomedusan does the axial portion of the modified tentacle retain so simply the character of the cartilaginoid axial tissue of a tentacle, and although in other Trachomedusæ an otolithic concretion is formed by the axial cells, whilst an otolith is most decidedly not formed in Limnocodium, yet in the essential character of the refringent body, and in the remarkable ectodermal capsule, Limnocodium agrees most pre-

cisely with the Trachomedusæ, such as Rhopalonema, described by the Hertwigs. It appears that in Rhopalonema and other Trachomedusæ, the marginal body originates as a small tentacle-like outgrowth with endodermal axis. Whilst the endodermal axis undergoes modifications, an ectodermal upgrowth (of the abumbral velar surface) occurs all round the little tentacle so as to form a sort of investing vesicle, which at first is open, but in the course of development closes up. The little tentacle is thus enclosed in a spherical sac or capsule.

Precisely the same history attaches to the tentacle-like commencement of the refringent body of Limnocodium, excepting that the capsule commences somewhat early, and that the whole structure, both tentacle-body and capsule, instead of standing out freely on the abumbral surface of the velar insertion, keeps a special course of direction of growth, creeping so to speak between the two layers of the velum instead of pushing outwards at right angles to the abumbral surface.

There can, it seems to me, be no doubt that the refringent body of Limnocodium is identical with the " free auditory bulbs" (freie Hörkölbchen) of Trachyline Medusæ, in which phase the so called auditory organ of Trachomedusæ makes its first appearance. The "tubular capsules" or "velar centrifugal canals," which enclose the refringent bulbs of Limnocodium, are identical with the at first open and subsequently closed auditory sacs of ectodermal origin, which grow up around the Hörkölbchen of Trachynemidæ, Olindiadæ, and Geryonidæ.

I have observed the development of true tentacles in the new Medusa, and may state that in them the endodermal axis at once begins to assume its special character, which prevents any possibility of confusion (apart from their different positions) between the true tentacle and the tentacular " auditory " bulb.

Until it is shown (as it appears to me very possibly may some day be shown) that some Leptomedusæ have their marginal bodies formed by modified tentacles, as in Trachomedusæ and Narcomedusæ, and not purely and simply from ectodermal cysts, as the Hertwigs have described, the features which I have shown to characterise the development and essential structure of the marginal bodies of Limnocodium will render it necessary, even apart from those features of the tentacles and marginal ring above described, to associate that form, not with Leptomedusæ, but with the Trachomedusæ, as I pointed out when I gave to it the name Craspedacustes.

Embryonic condition of Limnocodium.—Not the least astonishing fact about the new fresh-water Medusa is the immense preponderance of males. Among more than fifty specimens which

I have examined, I have not found one female.   Yet females, or egg-bearing forms, are there.

When I first received specimens of the Medusa from Mr. Sowerby, he imparted to me his conviction that young Medusæ were being hatched in the glass jar in his sitting-room, which contained a number of full-grown specimens.   He inferred this from the fact that among these large specimens which he had introduced into the jar a number of minute specimens made their appearance after the lapse of three or four days.   On a subsequent occasion Mr. Sowerby again drew my attention to these minute Medusæ, and enabled me to examine a number of them.

Had I been preoccupied with the notion that Limnocodium must have a hydriform trophosome, I might perhaps have neglected the opportunity of examining these minute forms.   But as I had come to the conclusion, on anatomical grounds, that Limnocodium is one of the Trachomedusæ, I was quite prepared, when Mr. Sowerby for the second time mentioned these minute Medusæ, to find very minute Limnocodia hatched from eggs in the vessels in which the adults were kept.

The youngest specimen which I have at present examined measured only one thirtieth of an inch in diameter, and I have examined others very little larger.

They agree in a very striking way with the embryo of *Geryonia hastata,* figured by Metschnikow in the 'Zeitsch. für wiss. Zoologie,' Bd. xxiv, pl. ii, figs. 13, 15.   There is no possibility of doubting that these embryos were developed from eggs, just as are those of Geryonia, Cunina, and Æginopsis.

The smallest embryo (woodcut, fig. 4) was of a slightly depressed, nearly spherical, form.   It exhibited that very striking separation of the ectoderm from the endoderm layer which is seen in the similar embryo of Geryonia.   Four perradial tentacles, very short and stump-like, were sprouting at a little distance from one depressed pole of the sphere.   Between these, rudiments of four others were seen.   The external ectodermal layer was unbroken and not invaginated at any part of the sphere.   Yet within could be seen the subumbrellar cavity, the manubrium with its mouth, the quadrangular stomach, and the four radiating canals.

The ectoderm was, it is very strange to say, continued across the mouth of the umbrella, so as to close in the subumbrellar space.   Were this covering to be ruptured centrally the subumbrellar cavity would be found ready formed, with dependent manubrium and mouth.   The ectodermal lid thus centrally perforated would very readily become the velum.

I am at this moment unable to say whether this is the mode of development, since in specimens a little larger with

eight tentacles, fig. 5, the præumbral lid was still imperforate.[1]
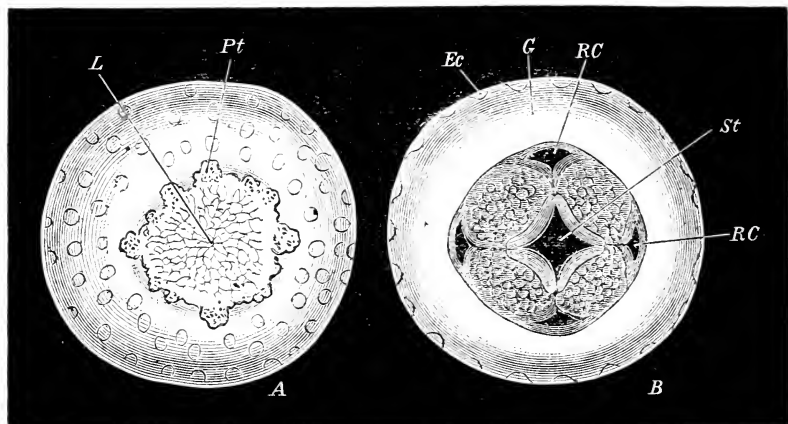
FIG. 4.—Embryo of *Limnocodium Sowerbii*, $\frac{1}{30}$th of an inch in diameter. *A*. Surface view of oral pole. *B*. Optical section of same specimen in a plane at right angles to the oro-apical axis. *Pt*. Per-radial tentacle. *L*. Præumbral lid. *RC*. Radial canal. *St*. Stomach. *Ec*. Ectoderm. *G*. Jelly of the disc.
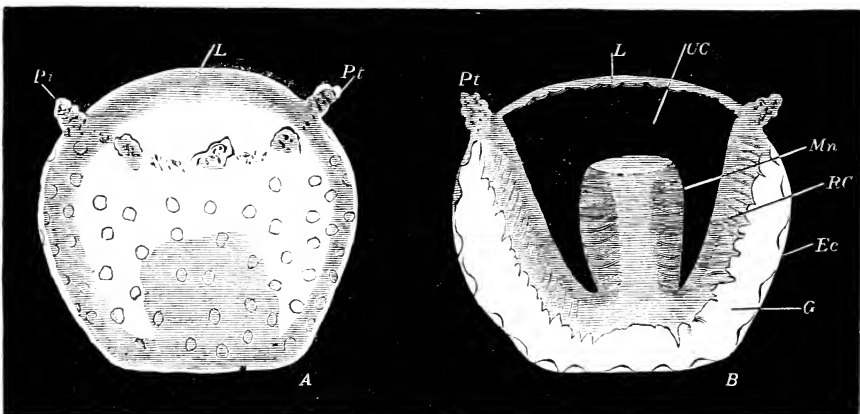
FIG. 5.—Embryo of *Limnocodium Sowerbii*, a very little more advanced. *A*. Surface lateral view. *B*. Optical section of the same specimen in a plane including the oro-apical axis. *Pt*. Per-radial tentacles. *L*. Præumbral lid. *UC*. Sub-umbrella cavity. *Mn*. Manubrium. *RC*. Radial canal. *Ec*. Ectoderm. *G*. Jelly-like substance of the disc.

[1] Whilst this is passing through the press I have ascertained that such is the development of the velum.

The movements of the embryo in this phase are active enough. A frequent pulsation of the subumbrellar musculature is seen, which alters but little the surface outline of the spherical embryo. The spherical form *is*, however, slowly altered, and occasionally the embryos become *much* elongated parallel to the oro-apical axis.

I defer any further description of the development of Limnocodium until I can submit the more complete drawings of the stages which I have already recorded and of others for which I am now in search.

Intra-cellular digestion in Limnocodium.—The exceedingly important fact that some of the Cœlentera, and lower kinds of worms, digest their solid food by the inception of the solid food-particles into the substance of endodermal cells, each endodermal cell behaving as an Amœba, has now been fairly established by the observations of Allman on Myriothela, of Metschnikoff on Turbellarians, and of T. J. Parker on the common Hydra.

Limnocodium exhibits this mode of digestion in the most striking and obvious manner. I have prepared drawings of the endodermal cells of the stomach, showing their amœboid character, and showing further the presence of such food-bodies as Protococci, Diatoms, and Euglenæ, in various stages of digestion, *within* the protoplasm of single cells, and of aggregated groups of such cells. These were observed in and drawn from living specimens of Limnocodium with a magnifying power of 800 diameters.

On the Development of the Structure known as the 'Glo-
merulus of the Head-Kidney' in the Chick.[1] By Adam Sedg-
wick, B.A.—In a paper by Mr. Balfour and myself in the 'Quart.
Journ. of Micr. Science,' vol. xix, describing the development of
what we believed to be a rudimentary head-kidney in the chick,
we drew attention to a structure which so closely resembled the
glomerulus of the head-kidney of the Icthyopsida that we iden-
tified it as an homologous structure.

Gasser[2] has also independently discovered and similarly iden-
tified this structure.

In the paper just referred to no attempt was made to trace
the development of this glomerulus, but it was merely described
as it appeared at its time of greatest development.

The following description is taken from that paper :

"In the chick the glomerulus is paired and consists of a vas-
cular outgrowth or ridge projecting into the body cavity on each
side at the root of the mesentery. It extends from the anterior
end of the Wolffian body to the point where the foremost open·
ing of the head-kidney commences. We have found it at a
period slightly earlier than that of the first development of the
head-kidney. In the interior of this body is seen a stroma with
numerous vascular channels and blood-corpuscles, and a vas-
cular connection is apparently becoming established, if it is not
so already, between the glomerulus and the aorta. The stalk
connecting the glomerulus with the attachment of the mesentery
varies in thickness in different sections, but we believe that the
glomerulus is continued unbroken throughout the very consider-
able region through which it extends. This point is, however,
difficult to make sure of, owing to the facility with which the
glomerulus breaks away. At the stage we are describing no
true Malpighian bodies are present in the part of the Wolffian
body on the same level with the anterior end of the glomerulus,
but the Wolffian body merely consists of the Wolffian duct. At

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[1] Read before the Cambridge Philosophical Society.
[2] 'Sitzungsberichte der Gesellschaft zur Beförd d. gesam. Naturwiss.,'
No. 5, 1879.

the level of the posterior part of the glomerulus this is no longer the case, but here a regular series of primary Malpighian bodies is present, and the glomerulus of the head-kidney may frequently be seen in the same section as a Malpighian body. In most sections the two bodies appear quite disconnected, but in those sections in which the glomerulus of the Malpighian body comes into view it is seen to be derived from the same formation as the glomerulus of the head-kidney."

The point which is left in doubt in the above description, viz. as to whether the glomerulus constitutes a continuous structure, is at once decided by a study of its development.

I may here state that it is not a continuous structure, but consists of a series of external glomeruli, each of which corresponds and is continuous with the glomeruli of the Malpighian bodies found in this part of the trunk.

I will commence the description of the development at the time when the segmental tubes have reached the stage of development figured by Kölliker[1] and myself.[2] At this stage each of them in the anterior region of the Wolffian body has the form of an S-shaped string, with a narrow opening into the body cavity, the lower limb of the S being formed by the intermediate cell mass, and the upper limb by a column of cells which connects the intermediate cell mass with the Wolffian duct.

In the region where each external glomerulus is afterwards found the openings into the body cavity, which are homologous with the peritoneal openings of the segmental tubes in Elasmobranchs, widen out very considerably, and a lumen is continued from them into the intermediate cell mass on the one hand, and on the other hand into the column of cells which forms the upper limb of the S and connects the intermediate cell mass with the Wolffian duct.[3]

That part of the segmental tube which will afterwards become a Malpighian body is therefore, in the region where an external glomerulus will subsequently be formed, connected with the body

[1] This may best be understood by examining fig. 11, Pl. XVII, in my paper already referred to ('Quart. Journ. of Micr. Science,' April, 1880). If the primary Wolffian tubule (wt'), here represented, were connected with the peritoneal epithelium at the point where the line from wt' cuts it, and it were open to the body cavity at that point, an appearance similar to that which I have attempted to describe would be obtained. Or perhaps a better idea of the structure may be obtained from fig. 6, Pl. XX, in Balfour's 'Monograph on the Development of Elasmobranch Fishes.' If st were very short and wide, so that mg were widely open to the body cavity, the figure would resemble a developing Wolffian tubule in this anterior part of the chick's Wolffian body.

[2] 'Entwicklungsgeschichte des Menschen u. der höheren Thiere,' p. 201, 2nd ed.

[3] 'Quart. Journ. Mic. Sci.,' April, 1880, Pl. XVII, fig. 1.

cavity by a short tube.   This tube rapidly widens out, especially anteriorly, to such an extent that it soon appears as a shallow bay in the body cavity.   Thus each opening at this stage forms a bay, wide and shallow anteriorly, becoming deeper and narrower as we pass backwards, until finally behind it is separated from the body cavity altogether, and there is seen in section a Malpighian capsule precisely resembling a developing Malpighian capsule in the hinder region of the Wolffian body.[1] In this bay and in the small part behind continuous with the bay, but separated from the body cavity, which are together serially homologous with a Malpighian capsule and the funnel leading from it into the body cavity, a small glomerulus soon appears attached to the dorsal wall.   The glomerulus increases in size, and the bay anteriorly widens out very much, while behind it remains deep, and finally passes into the closed posterior portion.   The glomerulus fills up this passage, which clearly runs obliquely backwards and dorsalwards, and eventually, as far as I can ascertain, the opening becomes completely closed, the epithelium on the external glomerulus being no longer continued through the opening on to the internal glomerulus.

The external glomerulus, then, in the chick which has hitherto been known as the glomerulus of the head-kidney, is nothing more than a series of glomeruli of primary Malpighian bodies projecting through the wide openings of the segmental tubes into the body cavity.   Their extreme antero-posterior extension may be said to be within the ninth and thirteenth segments.

In the chick the primary segmental tubes corresponding to these external glomeruli are apparently never fully developed.

I may mention that the external glomeruli are present in greater numbers, and attain a greater development in the duck than in the chick.

I defer the details and all discussion of this extraordinary and unexpected development until I am able to publish a fuller paper with figures.

Bacterium Anthracis.—Professor Greenfield, of the Brown Institution, has recently made the following report to the Royal Agricultural Society :

In my former report, published in the last volume of the Society's journal, I gave the results of my experiments so far as they were completed to the middle of February.

It may be remembered that the chief results of the experiments, briefly stated, were—

(1) That splenic fever may be transmitted to a bovine

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[1] Loc. cit., fig. 11.

animal by direct inoculation from a Rodent; that the disease thus given, though severe and possibly fatal, is not usually so, and that the modified attack of the disease confers a certain degree of protection from subsequent attacks communicated in the same way; so far, at any rate, as the experiments had been carried.

(2) I showed also that the fungus which constitutes the essential contagium, when grown in successive generations in a cultivating fluid, was still capable of giving rise to the disease, being in one case fatal to a cow in the first generation, in another fatal to a sheep in the fourth generation. My experiments also showed that an attack thus communicated, causing severe symptoms, appeared to be equally protective against results from future inoculations with the disease given directly from the guinea-pig.

I pointed out that, although I had applied as severe tests as were at my command, to ascertain the degree of protection conferred, it was yet desirable to perform the more crucial test of direct contagion from another bovine animal; and that, if the experiments were successful, the precise method of performing the protective inoculation, and the durability of the protection, would require further investigation. Having regard, also, to the small number of animals as yet experimented on, it would be desirable to repeat similar trials on others.

Keeping these points in view, I have continued the experiments, and have inoculated two other animals with similar results. In one case also, which had just recovered from the first attack at the time of my report, I have ascertained by further inoculations that protection had been conferred. Thus in all, I have added three more to the cases of success by this method. In one of these, the disease was given by the fourth generation of the cultivated fungus, and the symptoms were severe.

So far as concerns the crucial experiment, that of exposure to direct contagion from another bovine animal, I have not as yet had any opportunity of making it, and am still awaiting the occurrence of some outbreak, of which I hope we may receive early information. But I have just received some material from other animals which is known to be highly infectious, and am about to make experiments with it.

Since my previous report, another very interesting and equally important point has become clear, which may I hope prove of great value in future. It is that, when the virus of the disease (the fungoid organism known as *Bacillus*

anthracis) is artificially cultivated in an indifferent fluid, by the method of successive generations, which I have described in my report, each successive generation becomes less active than its predecessor, and when inoculated acts not only with less intensity, but more gradually, and often in a somewhat different manner. This modification takes place to such a degree that when the cultivation has been carried to the fourteenth or fifteenth generation, it may be introduced with impunity into the system of a mouse, which is one of the animals most susceptible to the poison.

Apart from its scientific interest, this fact will doubtless prove to be of practical value, for by its means it will be possible to obtain a virus of sufficient activity to produce an attack of the disease which shall be protective, but not of sufficient severity to be dangerous, or in any way injurious to the animal inoculated.

With regard to any apprehended ill-effect upon the animals thus inoculated, I may say that the cows which we have used have thriven remarkably well, and none so well as that which has been most severely tested.

I hope in a future report to give the details of these investigations, which have necessarily been extensive and complicated.

I venture, therefore, to urge upon the Society the importance of continuing these experiments, so as to bring them to a complete and decisive result. In order to do this, further outlay in the purchase and keep of animals, and other expenses, will be necessary, which will involve the renewal of the grant for the ensuing six months.

Dr. Carl Rabl on the Pedicle of Invagination in Pulmonate Gastropoda.—Dr. Rabl has renewed his investigations on the embryology of Planorbis, and has arrived at a result which brings his observations of fact and my own (published in this journal in 1874) into close agreement on a matter of considerable importance concerning which he was at first led to differ from me.

He writes relatively to the question of the existence of a " pedicle of invagination " as follows (dated June 4th):

" The youngest embryos, of which I have cut sections in order to determine the above question, were little older than those drawn by me in Plate XXXII, fig. 20, of my Memoir. Although by observation of the embryos as whole objects I could see nothing which could lead to the inference of the existence of a pedicle of invagination, yet I convinced myself by the aid of sections that a cylindrical solid cord

exists which leads from the cavity of the mid-gut to the integument. This cord consists of small endoderm-cells, rich in granules, and the ectoderm-cells of the spot where it comes into contact with the integument are somewhat longer and poorer in granules than the neighbouring cells. There is, however, no trace to be seen of an orifice of invagination.

" In sections of somewhat older embryos one can observe that the cord becomes hollowed out, the hollowing process taking origin from the cavity of the mid-gut, and it is easy enough to convince oneself that it gives rise to the terminal gut. Accordingly I am in the pleasant position of being able to confirm your statements, in so far as that a cord exists which stretches from the integument to the gut, but, on the other hand, I consider that my view is confirmed, that the terminal gut is a derivative of the mid-gut.

" Your interpretation of this cord as a ' pedicle of invagination ' I am not able now, any more than before, to accept."

Dr. Rabl will explain his views and observations at greater length in Professor Gegenbaur's ' Morph. Jahrbuch.'

E. RAY LANKESTER.

Development of Muscular Tissue from Epiblast in the Mammalia.—According to recent observations of Ranvier this extremely important and unexpected fact has been established by him in regard to the muscular coat of the sweat-glands. The reader is referred to Ranvier's notice of his discovery in the ' Comptes Rendus of the Acad. of Sciences of Paris,' Dec. 29th, 1879.

PROCEEDINGS OF SOCIETIES.

Chroococcaceous alga from Leicester, exhibited.—Mr. Archer exhibited examples from a small collection of a Chroococcaceous algal form, kindly forwarded him by Mr. Holmes, of London, which had occurred in considerable quantities in the water supply of the town of Leicester. This, regarded from what might be called a morphological point of view, and taken as it stood, would be referable to the genus Cœlosphærium, *Näg.*, but it was clearly not identical with the common *Cœlosphærium Kützingianum*, Näg., of which Mr. Archer exhibited some examples for comparison and illustration. He might mention that the latter form, when taken from running water, as these examples were, seems to possess notably smaller constituent cells than in the form taken from standing pools. In *C. Kützingianum*, however, the individual cells are globose. In Mr. Holmes' plant they were elongate, considerably larger, of a different tint and internal appearance. The fission of the cells took place in the direction of the longer axis, that is, radially, as regards the aggregate group or rotund colonies, whereby the constituent cells came to stand themselves radially, that is, pointing to the common centre. It may be a question, indeed, how far Cœlosphærium can be correctly regarded as a generic type ; for instance, the clearly allied form referred to Bacterium by Lankester, his *B. rubescens*, would appear to temporarily put on a Cœlosphærium-like condition—nay, also a Merismopædia-like, a Clathrocystis-like, and other phases. Since arrival many of the " Cœlosphærium " aggregations had become broken up the cells, still living and subdividing, the fission still in the longitudinal direction ; in that state the as yet not disconnected pairs of cells called to mind the appearance of a pair of cotyledons of some seed—say a bean—opened, and as yet held together by the embryo. Of course, this plant possessed considerable affinity to such as *Microspora (Clathrocystis) æruginosa*, agreeing, too, with it, as would appear, in its copious occurrence when met with ; but the elongate form of the cells and different tint were striking. A very slight pressure was sufficient to drive away from a colony a number of the constituent cells, and then, or when apparently normally broken up, of course the separated cells might pass for a species of Synechococcus (where the line of fission, however, is transverse), but just as well the same might be said for the separated cells of a Clathrocystis, which might pass for, say, *Chroococcus minor*. It does not seem at all unlikely that under some different impulses the vegetating cells of this group may

take on different modes of combination, and assume arrangements in themselves as yet supposed to be of generic import. Here, however, the cells themselves seemed to present an aspect unusual; if one could judge correctly, they seemed to offer a slightly tapering figure, but by no means comparable to that of the related *Gomphosphæria aponina*, in which the cells are almost peg-top shaped, or, when partially divided, " heart-shaped ; " but the present plant agrees with that in the longitudinal direction of the line of self-division. Probably a characteristic connected with their slightly unequally-ended figure was that the last point of connection between the two young cells not yet disjunct was not at the centre but notably nearer one end, thus increasing the resemblance (alluded to) to the opened but not sundered pair of cotyledons of a bean. In this plant the internal arrangement of the little granular and darkish masses of endochrome seemed peculiar.

Grains of Ruby in Australian sand.—Dr. Frazer showed some fine sand from Australia, taken from a river bed, many of the grains of which were of ruby, very pretty to look at under a low power, but far too small to be of any value. The sample had been given to him by the Rev. Mr. Whitmee.

Modifications of structure presented in the cross sections of the spines of Goniocidaris caniculatus, G. tubaria, and G. geranoides.—Prof. Mackintosh exhibited cross-sections of the spines of *Goniocidaris canicalatus*, A. Agassiz, *G. tubaria*, Lamk., and *G. geraniodes*, Lamk., and called attention to the interesting modification of the structure to be observed in them. In all three the central parts of the spine corresponded with the typical Cidarid arrangement. In the first named the peripheral crust was also sufficiently normal, there being no extensions of the intermediate part of the spine ; but in *G. geranioides*, as is well known, instead of the usual shell perforated by fine radiating tubules, there was an investment of reticulated tissue of the common echinoid type, supported by numerous extensions outwards of the intermediate parts of the spine tissue. (For figures of this form see Agassiz, ' Revision of the Echini,' pl. xxx, and Mackintosh, ' Trans. Roy. Irish Acad.,' vol. xvii, pl. ix.) In *G. tubaria* was seen the connecting form between the two extremes, for there the crust is still more or less reticulated, but the projections into it are few and comparatively short. Prof. Mackintosh was indebted for the specimens, from which the sections had been made, to Prof. A. Agassiz, of the Harvard University.

Minute quasi-parasitic Callithamnion on Lomentaria articulata.—Dr. E. Perceval Wright exhibited some small morsels of the young fronds of *Lomentaria articulata*, from the surface of which were to be seen in many stages of growth a very minute quasi-parasitic species of Callithamnion. It first made its appearance as a single filament of four or five cells, protruding from some of the outer cells of *L. articulata ;* this soon branched,

and in process of time the little plant reached dimensions just visible to the unassisted vision; at this stage its tetraspores were found, but no trace was detected of either carpogonia or antheridia. There were great difficulties in the way of referring this plant to any of Reinsch's species, many of which were indeed still without specific names. It had been found pretty common on *L. articulata*, collected about Howth during the spring of 1879, and doubtless would turn up again, when perhaps its true fruits might be discovered, and with them a more perfect account laid before the club.

Branching of the staminal hairs of Tradescantia. — Prof. M'Nab drew attention to a singular case of branching hairs from the stamens of *Tradescantia virginica*. The penultimate cell gave off at its base a branch consisting of two cells, the axis of growth of the branch being downwards and backwards. This form and three other hairs, each with a branch consisting of a single lateral cell, were all obtained from an unopened flowerbud, from a cut flower-stem, which had been some time in water, and therefore under abnormal conditions.

December, 1879.

Cosmarium isthmochondrum, Nordst., *new to Ireland, exhibited.* —Mr. Archer showed, new to Ireland, *Cosmarium isthmochondrum*, Nordst., from Connemara, and, similarly to Nordstedt's own example, as he states, in company with *Cosmarium quinarium*. Both these are very rare species, but when met with sometimes occurring in quantity. As Herr Nordstedt remarks, these two species might possibly be confounded, but Mr. Archer thought, if their distinction were but once properly appreciated, such a mistake could not occur on a second occasion, for it needed only a little careful observation to see their striking and strong points of difference; even with a low power, when one has got a good grasp of the outline of each, Mr. Archer thought they might be very readily discriminated. There could be no doubt but that *Cosmarium isthmochondrum* is a very good and distinct species.

Parasitic Florideous alga in Plocamium coccineum, exhibited. —Dr. E. Perceval Wright exhibited some filaments of a Florideous alga, which he had found in the interior of the main portion of the frond of *Plocamium coccineum*. These filaments were of a distinctly red colour, were frequently branched, and apparently grew up within the cell tissue of the Plocamium. Dr. Wright had found them in November, 1878, and was again reminded of them by the appearance, in the 'Botanische Zeitung' for 17th January, 1879, of a paper by Reinsch, on "Entozoische Pflanzenparasiten," in the plate accompanying which, fig. B, an almost identical form is figured. Unfortunately though watched from time to time, nothing had yet transpired to throw any additional light on their life history.

MEMOIRS.

LARVAL FORMS: *their* NATURE, ORIGIN, *and* AFFINITIES.
By F. M. BALFOUR, M.A., F.R.S., Fellow of Trinity
College, Cambridge.

Preliminary considerations.—Animals either (1) undergo
the whole of their early development in the egg or within
the body of the parent, and are hatched in a condition closely
resembling the adult; or else (2) they are born in a condi-
tion differing to a greater or less extent from the adult.
When born in the latter condition they are known as
larvæ, till they have approximately acquired the adult
characters of the species. There are no questions which are
of greater importance for the embryologist than those which
concern the nature of the secondary changes likely to occur
in the fœtal or in the larval state; since it is on the answer
to such questions that our knowledge of the extent to which
a record of the ancestral history may be expected to be pre-
served in development depends. The principles which govern
the perpetuation of variations which occur in either the larval
or the fœtal state are the same as those for the adult state.
Variations favorable to the survival of the species are equally
likely to be perpetuated, at whatever period of life they occur,
prior to the loss of the reproductive powers. The possible
nature and extent of the secondary changes which may have
occurred in the developmental history of forms, which have
either a long larval existence, or which are born in a nearly
complete condition, is primarily determined by the nature
of the favorable variations which can occur in each case.

Where the development is a fœtal one, the favorable
variations which can most easily occur are—(1) abbrevia-
tions, (2) an increase in the amount of food-yolk stored up for
the use of the developing embryo. Abbreviations take place
because direct development is always simpler, and therefore

more advantageous; and, owing to the fact of the fœtus not
being required to lead an independent existence till birth,
and of its being in the meantime nourished by food-yolk,
or directly by the parent, there are no physiological
causes to prevent the characters of any stage of the develop-
ment, which are only of functional importance to a *free
larva*, from disappearing from the developmental history.
All external organs of locomotion and nutrition will, for this
reason, obviously have a tendency to disappear or to be reduced
in fœtal developments; and a little consideration will show
that the ancestral stages in the development of the nervous
and muscular systems, organs of sense, and digestive system
will be liable to drop out or be modified, when a simplifica-
tion can thereby be affected.   The circulatory and excretory
systems will not be modified to the same extent, because both
of them are usually functional during fœtal life.

The mechanical effects of food-yolk are very considerable,
and numerous instances of their influence will be found in
my treatise on 'Comparative Embryology.'   They mainly
affect the early stages of development, *i.e.* the form of the
gastrula, &c.

The favorable variations which can occur in the free
larva are much less limited than those which can occur in the
fœtus.   Secondary characters are therefore very numerous
in larvæ, and there may even be larvæ with secondary
characters only, as, for instance, the larvæ of insects.

In spite of the liability of larvæ to acquire secondary
characters, there is a powerful counter-balancing influence
tending towards the preservation of ancestral characters, in
that larvæ are necessarily compelled at all stages of their
growth to retain in a functional state such systems of organs,
at any rate, as are essential for a free and independent
existence.   It thus comes about that in spite of the many
causes tending to produce secondary changes in larvæ, there
is always a better chance of their repeating, in an unabbre-
viated form, their ancestral history than is the case with
embryos, which undergo their development within the egg.

It may be further noted as a fact which favours the re-
lative retention by larvæ of ancestral characters, that a
secondary larval stage is less likely to be repeated in de-
velopment than an ancestral stage, because there is always
a strong tendency for the former, which is a secondarily inter-
calated link in the chain of development, to drop out by the
occurrence of a *reversion* to the original type of development.

The relative chances of the ancestral history being pre-
served in the fœtus or the larva may be summed up in the

following way :—There is a greater chance of the ancestral history being lost in forms which develop in the egg ; and masked in those which are hatched as larvæ.

The evidence from existing forms undoubtedly confirms the *a priori* considerations just urged.[1] This is well shown by a study of the development of *Echinodermata, Nemertea, Mollusca, Crustacea,* and *Tunicata.* The free larvæ of the four first groups are more similar amongst themselves than the embryos which develop directly, and since this similarity cannot be supposed to be due to the larvæ having been modified by living under precisely similar conditions, it must be due to their retaining common ancestral characters. In the case of the *Tunicata* the free larvæ retain much more completely than the embryos certain characters which are known to have been ancestral.

Types of Larvæ.—Although there is no reason to suppose that all larval forms are ancestral, yet it seems reasonable to anticipate that a certain number of the known types of larvæ should resemble the ancestors of the more important phyla of the animal kingdom.

Before examining in detail the claims of various larvæ to such a character, it is necessary to consider somewhat more at length the kind of variations which are most likely to occur in larval forms.

It is probable *a priori* that there are two kinds of larval forms, which may be distinguished as primary and secondary larval forms. Primary larval forms are more or less modified ancestral forms, which have continued uninterruptedly to develop as free larvæ from the time when they constituted the adult form of the species. Secondary larval forms are those which have become introduced into the ontogeny of species, the young of which were originally hatched with all the characters of the adult ; but which, owing to the loss of food-yolk in the egg, or some other cause, have become hatched at an earlier period. Such secondary larval forms may resemble the primary larval forms in cases where the ancestral characters were retained by the embryo in its development within the egg ; but in other instances the characters they have are probably entirely adaptive.

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[1] It has long been known that land and freshwater forms develop without a metamorphosis much more frequently than marine forms. This is probably to be explained by the fact that there is not the same possibility of a land or freshwater species extending itself over a wide area by the agency of free larvæ, and there is, therefore, much less advantage in the existence of such larvæ ; while the fact of such larvæ being more liable to be preyed upon than eggs, which are either concealed or carried about by the parent, might render it absolutely disadvantageous for a species to have such larvæ.

Causes tending to produce secondary changes in larvæ.—
The ways in which natural selection can act on larvæ may
probably be divided more or less artificially into two classes.

1. The changes in development necessarily produced by
the existence of a larval stage.

2. The adaptive changes in a larva acquired in the ordi-
nary course of the struggle for existence.

The changes which come under the first head consist
essentially in a displacement in the order of deve-
lopment of certain organs. There is always a tendency in
development to throw back the differentiation of the embry-
onic cells into definite tissues till as late a date as possible.
This takes place in order to enable the changes of form,
which every organ undergoes in repeating even in an abbre-
viated way its phylogenetic history, to be effected with the
least expenditure of energy. Owing to this tendency it
comes about that when an organism is hatched as a larva
many of the organs are still in an undifferentiated state,
although the ancestral form which this larva represents
had all its organs fully differentiated. In order, however,
that the larva may be enabled to exist as an independent
organism certain sets of organs, *e.g.* the muscular, nervous,
and digestive systems, have to be histologically differentiated.
If the period of hatching becomes earlier, an earlier differen-
tiation of certain organs is a necessary consequence; and in
almost all cases the existence of a larval stage causes a
displacement in order of development of organs, the com-
plete differentiation of many organs being retarded relatively
to the muscular, nervous, and digestive systems.

The possible changes under the second head appear to
be unlimited. There is, so far as I see, no possible reason
why an indefinite number of organs should not be developed
in larvæ to protect them from their enemies, and to enable
them to compete with larvæ of other species, and so on. The
only limit to such development appears to be the shortness of
larval life, which is not likely to be prolonged, since, *ceteris
paribus,* the more quickly maturity is reached the better it is
for the species.

A very superficial examination of marine larvæ shows that
there are certain peculiarities common to most of them, and
it is important to determine how far such peculiarities are to
be regarded as adaptive. Almost all marine larvæ are provided
with well-developed organs of locomotion, and transparent
bodies. These two features are precisely those which it
is most essential for such larvæ to have. Organs of locomo-
tion are important, in order that larvæ may be scattered as

widely as possible, and so disseminate the species; and transparency is very important in rendering larvæ invisible, and so less liable to be preyed upon by their numerous enemies.[1]

These considerations, coupled with the fact that almost all free-swimming animals, which have not other special means of protection, are transparent, seem to show that, at all events, the transparency of larvæ is adaptive, and it is probable that organs of locomotion are in many cases specially developed, and not ancestral.

Various spinous processes on the larvæ of Crustacea and Teleostei are also examples of secondarily acquired protective organs.

These general considerations are sufficient to form a basis for the discussion of the characters of the known types of larvæ.

The following table contains a list of the more important of such larval forms :

DICYEMIDÆ.—The Infusoriform larva.

PORIFERA.—(a) The Amphiblastula larva (fig. 1), with one half of the body ciliated, and the other half without cilia ; (b) the oval uniformly ciliated larva, which may be either solid or have the form of a vesicle.

CŒLENTERATA.—The planula (fig. 2).

TURBELLARIA.—(a) The eight-lobed larva of Müller (fig. 9); (b) the larvæ of Götte and Metschnikoff, with some Pilidium characters.

NEMERTEA.—The Pilidium (fig. 8).

TREMATODA.—The Cercaria.

ROTIFERA.—The Trochosphere-like larvæ of Brachionus (fig. 3) and Lacinularia.

MOLLUSCA.—The Trochosphere larva (fig. 4), and the subsequent Veliger larva (fig. 5).

BRACHIOPODA.—The three-lobed larva, with a postoral ring of cilia (fig. 6).

POLYZOA.—A larval form with a single ciliated ring surrounding the mouth, and an aboral ciliated ring or disc (fig. 15).

CHÆTOPODA.—Various larval forms with many characters like those of Molluscan Trochosphere frequently with distinct transverse bands of cilia. They are classified as Atrochæ, Mesotrochæ, Telotrochæ (fig. 12 A and fig. 13), Polytrochæ, and Monotrochæ (fig. 12 B).

GEPHYREA NUDA.—Larval forms like those of preceding groups. A specially characteristic larva is that of Echiurus (fig. 14).

GEPHYREA TUBICOLA.—Actinotrocha (fig. 17); with a postoral ciliated ring of arms.

MYRIAPODA.—A functionally hexapodous larval form is common to all the Chilognatha.

INSECTA.—Various secondary larval forms.

CRUSTACEA.—The Nauplius and the Zoœa.

ECHINDERMATA.—The Auricularia (fig. 10 A), the Bipinnaria (fig. 10 B), and the Pluteus (fig. 11), and the transversely-ringed larvæ of Crinoidea

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[1] The phosphorescence of many larvæ is very peculiar. I should have anticipated that phosphorescence would have rendered them much more liable to be captured by the forms which feed upon them; and it is difficult to see of what advantage it can be to them.

(fig. 7). The Auricularia, the Bipinnaria, and the Pluteus can be reduced to a common type (fig. 18 c).

ENTEROPNEUSTA.—Tornaria (fig. 16).

UROCHORDA (TUNICATA).—The tadpole-like larva.

GANOIDEA.—A larva with adhesive disc with papillæ in front of the mouth.

ANUROUS AMPHIBIA.—The tadpole.

Of the larval forms included in the above list a certain number are clearly without affinities outside the group to which they belong. This is the case with the larvæ of the Myriapoda, the Crustacean larvæ, and with the larval forms of the Chordata. I do not propose to discuss the significance of these forms in the present essay.
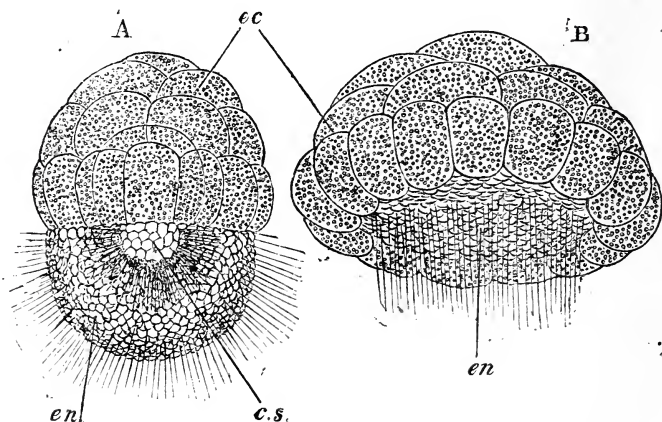
FIG. 1.—*Two free Stages in the development of Sycandra raphanus.* (After Schultze.) A. Amphiblastate stage. B. Stage after the ciliated cells have commenced to be invaginated. *c.s.* segmentation cavity; *ec.* granular epiblast cells; *en.* ciliated hypoblast cells.

There are, again, some larval forms which may possibly turn out hereafter to be of importance, but from which, in the present state of our knowledge, we cannot draw any conclusions. The infusoriform larva of the Dicyemidæ, and the Cercaria of the Trematodes are such forms.

The meaning of the Amphiblastula larva was discussed in a previous essay.

Excluding these and certain other forms, we have finally left for consideration the larvæ of the Cœlenterata, the Turbellaria, the Rotifera, the Nemertea the Mollusca, the Polyzoa, the Brachiopoda, the Chætopoda, the Gephyrea, the Echinodermata, and the Enteropneusta.

The larvæ of these forms can be divided into two groups.

The one group contains the larva of the Cœlenterata or Planula, the other group the larvæ of all the other forms.
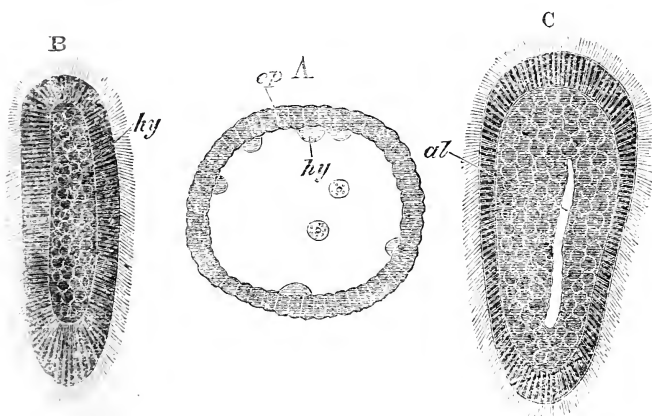
Fig. 2.—*Three larval Stages of Eucope Polystyla.* (After Kowalevsky. A. Blastophere stage with hypoblast spheres becoming budded into the central cavity. B. Planula stage with solid hypoblast. C. Planula stage with a gastric cavity. *ep.* epiblast; *hy.* hypoblast; *al.* gastric cavity.

The Planula (fig. 2) is characterised by its extreme simplicity. It is a two-layered organism, with a form varying from cylindrical to oval, and usually a radial symmetry. So long as it remains free it is not even provided with a mouth, and it is as yet uncertain whether or no the absence of a mouth is to be regarded as an ancestral character. The Planula is very probably the ancestral form of the Cœlenterata.

The larvæ of almost all the other groups, although they may be subdivided into a series of very distinct types, yet agree in the possession of certain characters.[1] There is a more or less dome-shaped dorsal surface and a flattened or concave ventral surface, containing the opening of the mouth, and usually extending posteriorly to the opening of the anus, when such is present.

The dorsal dome is continued in front of the mouth to form a *large præoral lobe*.

There is usually present at first a uniform covering of cilia; but in the later larval stages there are almost always formed definite bands or rings of long cilia, by which loco-

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[1] The larva of the Brachiopoda does not possess most of the characters mentioned below. It is probably all the same, a highly differentiated larval form belonging to this group.

motion is effected. These bands are often produced into
arm-like processes.
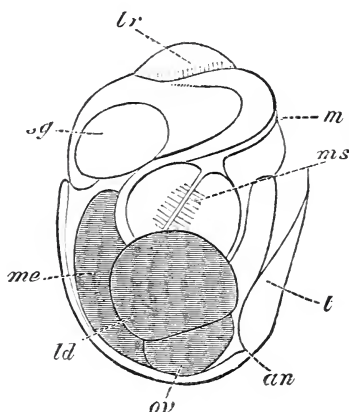
FIG. 3.—*Embryo of Brachionus urceolaris, shortly before it is hatched.*
(After Salensky.)   *m.* mouth; *ms.* masticatory apparatus; *me.* mesen-
teron; *an.* anus; *ld.* lateral gland; *ov.* ovary; *t.* tail, *i.e.* foot; *tr.*
trochal disc; *sg.* supra-œsophageal ganglion.

The alimentary canal has, typically, the form of a bent
tube with a ventral concavity, constituted (when an anus is
present) of three sections, viz. an œsophagus, a stomach, and
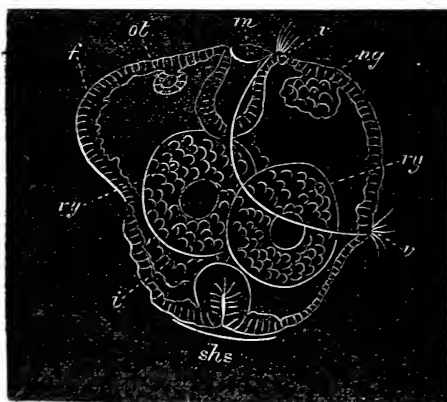
FIG. 4.—*Diagram of an Embryo of Pleurobranchidium.* (From Lankester.)
f. foot; *ot.* otocyst; *m.* mouth; *v.* velum; *ng.* nerve ganglion;
ry. residual yolk spheres; *shs.* shell-gland; *i.* intestine.

a rectum. The œsophagus and rectum are epiblastic in
origin, while the stomach is derived from the hypoblast.

To the above characters may be added a glass-like transparency; and the presence of a widish space, often traversed by contractile cells, between the alimentary tract and the body wall.

Considering the very profound differences which exist between many of these larvæ it may seem that the characters just enumerated are hardly sufficient to justify my grouping of them together. It is, however, to be borne in mind that my grounds for doing so depend quite as much upon the fact
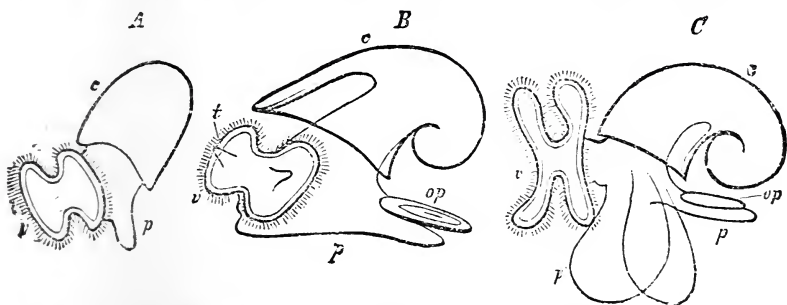
FIG. 5.—*Larvæ of Cephalophorous Mollusca in the veliger stage.* (From Gegenbaur.) A. and B. Earlier and later stage of Gasteropod. C. Pteropod (Cymbulia). *v.* velum; *c.* shell; *p.* foot; *op.* operculum; *t.* tentacle.

that they constitute a series without any great breaks in it, as upon the existence of characters common to the whole of them. It is also worth noting that most of the characters
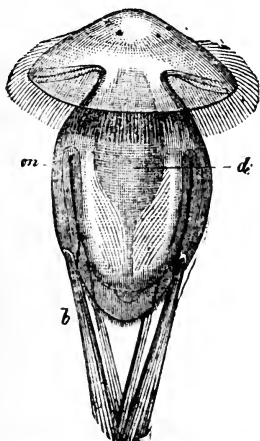
FIG. 6.—*Larva of Argiope.* (From Gegenbaur, after Kowalevsky.) *m.* mantle; *b.* setæ; *d.* archenteron.

which have been enumerated as common to the whole of these larvæ are not such secondary characters as (in accordance with the considerations used above) might be expected to arise from the fact of their being subjected to nearly similar conditions of life. Their transparency is, no doubt, such a secondary character, and it is not impossible that
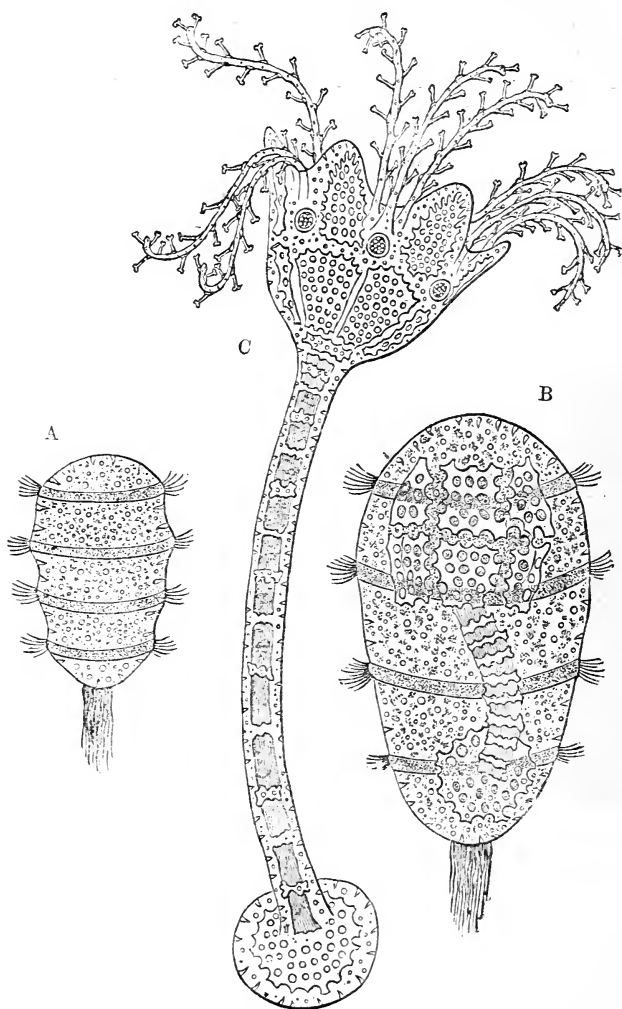
FIG. 7.—*Three Stages in the development of Antedon (Comatula)*. (From Lubbock, after Thompson.) A. Larva just hatched. B. Larva with rudiment of the calcareous plates. C. Pentacrinoid larva.

the existence of ciliated bands may be so also; but it is more probable that if, as I suppose, these larvæ reproduce the characters of some ancestral form, this form may have existed at a time when all marine animals were free-swimming, and that it may, therefore, have been provided with at least one ciliated band.

The detailed consideration of the characters of these larvæ, given below, supports this view.

This great class of larvæ may, as already stated, be divided into a series of minor subdivisions. These subdivisions are the following:

1. THE PILIDIUM GROUP.—This group is characterised by the mouth being situated nearly in the centre of the ventral surface, and by the absence of a proctodæum. It includes the Pilidium of the Nemertines (fig. 8), and the various larvæ of marine Dendrocœla (fig. 9). At the apex of the præoral lobe a thickening of epiblast may be present, from which (fig. 19) a contractile cord sometimes passes to the œsophagus.
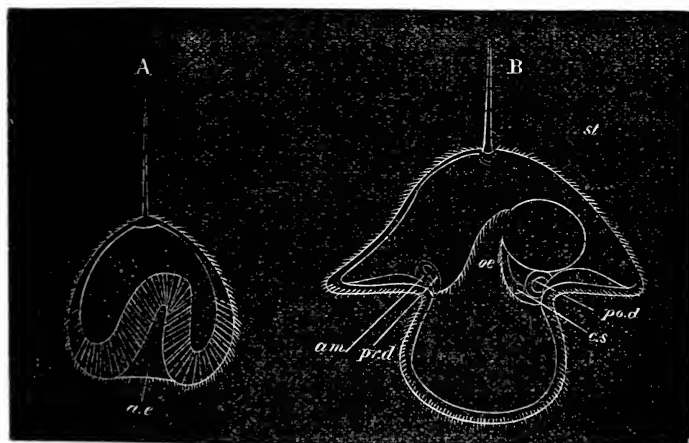
FIG 8.—*Two Stages in the development of Pilidium.* (After Metschnikoff.) *ae.* archenteron; *oe.* œsophagus; *st.* stomach; *am.* amnion; *pr.d.* prostomial disc; *po.d.* metastomial disc; *c.s.* cephalic sack.

2. THE ECHINODERM GROUP.—This group (figs. 10, 11, and 18 c) is characterised by the presence of a longitudinal postoral band of cilia, the absence of special sense organs in the præoral region, and the development of the body-cavity as an outgrowth of the alimentary tract. The three typical divisions of the alimentary tract are present, and there is a more

or less developed præoral lobe. This group only includes the larvæ of the Echinodermata.

3. THE TROCHOSPHERE GROUP.—This group (figs. 12, 13) is characterised by the presence of a præoral ring of long cilia, the region in front of which forms a great part of the præoral lobe. The mouth opens immediately behind the præoral ring of cilia, and there is very often a second ring of short cilia parallel to the main ring, immediately behind the mouth. The function of the ring of short cilia is nutritive, in that the cilia are employed in bringing food to the mouth; while the function of the main ring is locomotive. A perianal patch or ring of cilia is often present (fig. 12 A), and in many forms intermediate rings are developed between the præoral and perianal rings.

The præoral lobe is usually the seat of a special thickening of epiblast, which gives rise to the supra-œsophageal
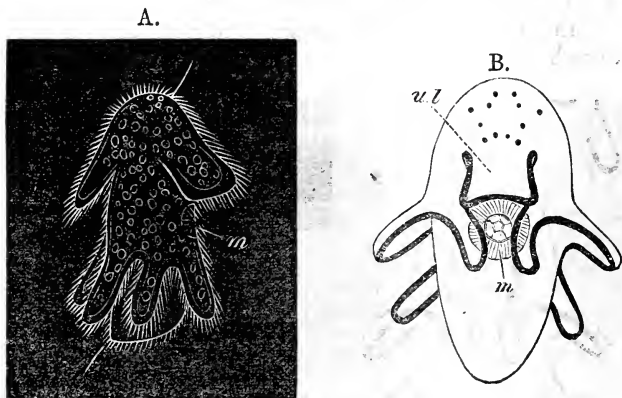
FIG. 9.—A. *Larva of Eurylepta auriculata immediately after hatching. Viewed from the side.* (After Hallez.) *m.* mouth. B. *Müller's Turbellarian Larva (probably Thysanozoon). Viewed from the ventral surface.* (After Müller.) The ciliated band is represented by the black line. *m.* mouth; *u.l.* upper lip.

ganglion of the adult. On this lobe optic organs are very often developed in connection with the supra-œsophageal ganglion, and a contractile band frequently passes from this region to the œsophagus.

The alimentary tract is formed of the three typical divisions.

The body-cavity does not develop directly as an outgrowth of the alimentary tract, though the process by which it originates is very probably secondarily modified from a pair of alimentary outgrowths.

Paired excretory organs opening to the exterior and into the body cavity are often present.
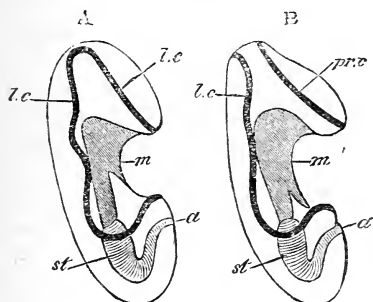
FIG. 10.—A. *The Larva of a Holothuroid.* B. *The Larva of an Asterias.* *m.* mouth; *st.* stomach; *a.* anus; *l.c.* primitive longitudinal ciliated band; *pr.c.* præoral ciliated band.

This type of larva is found in the Rotifera (fig. 3) (where it is preserved in the adult state), the Chætopoda, the Mollusca (fig. 4), the Gephyrea nuda (fig. 14), and the Polyzoa (fig. 15).[1]

4. TORNARIA.—This larva (fig. 16) is intermediate in most of its characters between the larvæ of the Echinodermata (more especially the Bipinnaria) and the Trochosphere. It agrees with Echinoderm larvæ in the possession of a longitudinal ciliated band (divided into a præoral and a postoral ring), and in the derivation of the body-cavity and water-vascular vesicle from alimentary diverticula; and it resembles the Trochosphere in the presence of sense organs

FIG. 11.—*A Larva of Strongylocentrus.* (From Agassiz.) *m.* mouth; *a.* anus; *o.* œsophagus; *d.* stomach; *c.* intestine; *v'.* and *v.* ciliated ridges; *w.* water-vascular tube; *r.* calcareous rods.

[1] For a discussion as to the structure of the Polyzoon larva, *vide* 'A Treatise on Comparative Embryology,' vol. i, p. 253.

on the præoral lobe, in the existence of a perianal ring of
cilia, and in the possession of a contractile band passing
from the præoral lobe to the œsophagus.

5. ACTINOTROCHA.—The remarkable larva of Phoronis,
(fig. 17) known as Actinotrocha, is characterised by the
presence of (1) a postoral and somewhat longitudinal ciliated
ring produced into tentacles, and (2) a perianal ring. It
is provided with a præoral lobe, and a terminal or some-
what dorsal anus.

6. The larva of the *Brachiopoda articulata* (fig. 6).

The relationships of the six types of larval forms just
characterised have been the subject of a considerable
amount of controversy, and the following suggestions on
the subject must be viewed as somewhat speculative. The
Pilidium type of larva is in some important respects less
highly differentiated than the larvæ of the five other groups.
It is, in the first place, without an anus; and there are no
grounds for supposing that the anus has become lost by
retrogressive changes. If for the moment it is granted that
the Pilidium larva represents more nearly than the larvæ of
the other groups the ancestral type of larva, what characters
are we led to assign to the ancestral form which this larva
repeats?
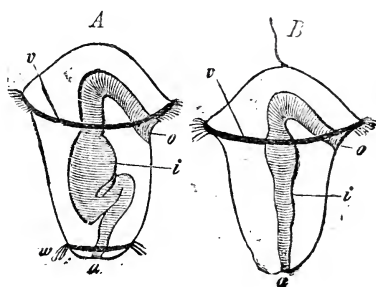
FIG. 12.—*Two Chætopod Larvæ.* (From Gegenbaur.) *o.* mouth; *i.* intes-
tine; *a.* anus; *v.* præoral ciliated band; *w.* perianal ciliated band.

In the first place, this ancestral form, of which fig. 18 A is
an ideal representation, would appear to have had a dome-
shaped body, with a flattened oral surface and a rounded
aboral surface. Its symmetry was radial, and in the centre
of the flattened oral surface was placed the mouth, and round
its edge was a ring of cilia. The passage of a Pilidium-
like larva into the vermiform bilateral Platyelminth form,
and therefore it may be presumed of the ancestral form which
this larva repeats, is effected by the larva becoming more
elongated, and by the region between the mouth and one

end of the body becoming the præoral region, and that
between the mouth and the opposite end developing into
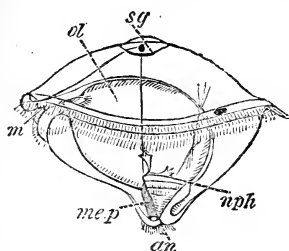
FIG. 13.—*Polygordius Larva.* (After Hatschek.) *m.* mouth; *sg.* supra-
œsophageal ganglion; *nph.* nephridion; *me.p.* mesoblastic band;
an. anus; *ol.* stomach.

the trunk, an anus becoming placed at the extremity of
the trunk in the higher forms.

If what has been so far postulated is correct, it is clear
that this primitive larval form bears a very close resemblance
to a simplified free-swimming Cœlenterate (Medusa), and that
the conversion of such a radiate form into the bilateral
took place, not by the elongation of the aboral surface, and
the formation of an anus there, but by the unequal elonga-
tion of the oral face, an anterior part forming a præoral
lobe, and a posterior part the trunk; while the aboral
surface became the dorsal surface.

This view fits in very well with the anatomical resem-
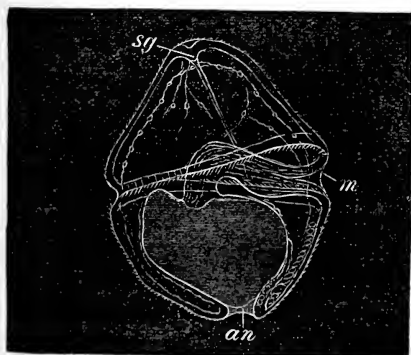
FIG. 14.—*Larva of Echiurus.* (After Salensky.) *m.* mouth; *an.* anus;
sg. supra-œsophageal ganglion (?).

blances between the Cœlenterata and the Turbellaria,[1] and

[1] *Vide* ' A Treatise on Comparative Embryology,' vol. i, p. 148 and 158.
In this connection attention may be called to *Coleoplana Metschnikowei*,

shows, if true, that the ventral and median position of the mouth in many Turbellaria is the primitive one.

The above suggestion as to the mode of passage from the radiate into the bilateral form differs entirely from t hat usually held. Lankester,[1] for instance, gives the follow ing account of this passage :
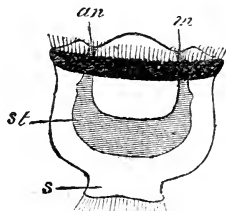
Fig. 15.—*Diagram of a Larva of the Polyzoa.* m. mouth; an. anus st. stomach ; s. ciliated disc.

"It has been recognised by various writers, but notably by Gegenbaur and Haeckel, that a condition of radiate symmetry must have preceded the condition of bilateral symmetry in animal evolution. The Diblastula may be conceived to have been at first absolutely spherical with spherical symmetry. The establishment of a mouth lead necessarily to the establishment of a structural axis passing through the mouth, around which axis the body was arranged with radial symmetry. This condition is more or less perfectly maintained by many Cœlenterates, and is reassumed by degradation of higher forms (Echinoderms, some Cirrhipedes, some Tunicates). The next step is the differentiation of an upper and a lower surface in relation to the horizontal position, with mouth placed anteriorly, assumed by the organism in locomotion. With the differentiation of superior and inferior surface, a right and a left side, complimentary one to the other, are necessarily also differentiated. Thus the organism becomes bilaterally symmetrical. The Cœlentera are not wanting in indications of this bilateral symmetry, but for all other higher groups of animals it is a fundamental character. Probably the development of a region in front of, and dorsal to the mouth, forming the *Prostomium,* was accomplished *pari passu* with the development of bilateral symmetry. In the radially

a form described by Kowalevsky, 'Zoologischer Anzeiger,' No. 52, p. 140, as being intermediate between the Ctenophora and the Turbellaria. There does not, however, appear to me to be sufficient evidence to prove that this form is not merely a creeping Ctenophor.

[1] 'Quart. Journ. of Micr. Science,' vol. xvii, pp. 422-3.

symmetrical Cœlentera we find very commonly a series of lobes of the body-wall or tentacles produced *equally*—with
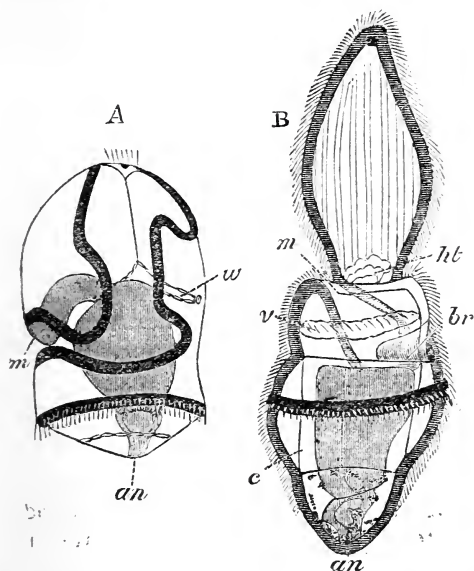
FIG. 16.—*Two Stages in the development of Tornaria.* (After Metschnikoff.) The black lines represent the ciliated bands. *m.* mouth; *an.* anus; *br.* branchial cleft; *ht.* heart; *c.* body-cavity between splanchnic and somatic mesoblast layers; *w.* so-called water-vascular vesicle; *v.* circular blood-vessel.

radial symmetry, that is to say—all round the mouth, the mouth terminating the main axis of the body—that is to say, the organism being ' telostomiate.' The later fundamental form, common to all animals above the Cœlentera, is attained by shifting what was the main axis of the body—so that it may be described now as the ' enteric' axis; whilst the new main axis, that parallel with the plane of progression, passes through the dorsal region of the body running obliquely in relation to the enteric axis. Only one lobe or outgrowth of those radially disposed in the telostomiate organisms now persists. This lobe lies dorsally to the mouth, and through it runs the new main axis. This lobe is the *Prostomium,* and all the organisms which thus develop a new main axis, oblique to the old main axis, may be called prostomiate."

It will be seen from this quotation that the aboral part of the body is supposed to elongate to form the trunk, while the præoral region is derived from one of the tentacles.

Before proceeding to further considerations as to the origin
of the Bilateralia, suggested by the Pilidium type of larva,
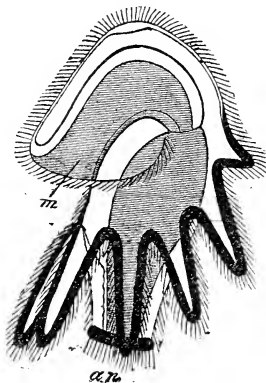
FIG. 17.—*Actinotrocha*.   (After Metschnikoff.)   *m*. mouth ; *an*. anus.

it is necessary to enter into a more detailed comparison be-
tween our larval forms.

A very superficial consideration of the characters of these
forms brings to light two important features in which they
differ, viz. :

(1) In the presence or absence of sense organs on the
præoral lobe.

(2) In the presence or absence of outgrowths from the
alimentary tract to form the body-cavity.

The larvæ of the Echinodermata and (?) Actinotrocha are
without sense organs in the præoral lobe, while the other
types of larvæ are provided with them. Alimentary diverticula
are characteristic of the larvæ of the Echinodermata and of
Tornaria.

If the conclusion already arrived at, to the effect that the
prototype of the six larval groups was descended from a
radiate ancestor, is correct, it appears to follow that the
nervous system, in so far as it was differentiated, had primi-
tively a radiate form, and it is also probably true that there
were alimentary diverticula in the form of radial canals, *two*
of which may have given origin to the paired diverticula
which become the body-cavity in such types as the Echino-
dermata.   If these two points are granted, the further con-
clusions seem to follow—(1) that the ganglion and sense
organs of the præoral lobe were secondary structures which
arose (perhaps as differentiations of the original circular
nerve-ring) after the assumption of a bilateral form ; and (2)
that the absence of these organs in the larvæ of the Echino-

dermata, and Actinotrocha (?), implies that these larvæ retain, so far, more primitive characters than the Pilidium. The same may be said of the alimentary diverticula. There are
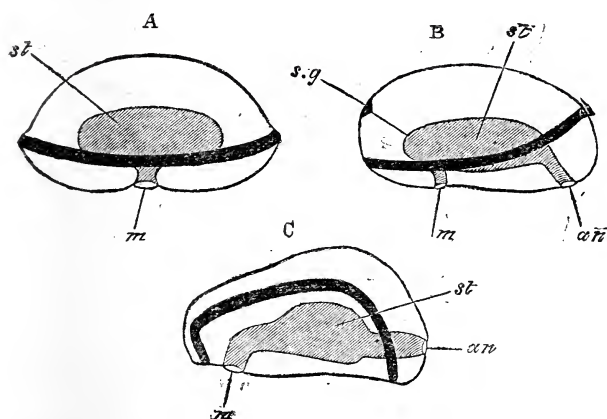
FIG. 18.—*Three Diagrams representing the ideal Evolution of various Larval forms.* A. Ideal ancestral larval form. B. Trochosphere larva. C. Echinoderm larva. *m.* mouth; *an.* anus; *st.* stomach; *s.g.* supra-œsophageal ganglion. The black lines represent the ciliated bands.

thus indications that in two important points the Echinoderm larvæ are more primitive than the Pilidium.

The above conclusions, with reference to the Pilidium and Echinoderms, involve some not inconsiderable difficulties, and suggest certain points for further discussion.

In the first place it is to be noted that the above speculations render it probable that the type of nervous system from which that found in the adults of the Echinodermata, Platyelminthes, Chætopoda, Mollusca, &c., is derived, was a circumoral ring, like that of Medusæ, with which radially arranged sense organs may have been connected; and that in the Echinodermata *this form of nervous system has been retained,* while in the other types it has been modified by the anterior part, having given rise to supra-œsophageal ganglia and organs of vision; which were formed owing to the assumption of a bilateral symmetry, and the consequent necessity for the sense organs to be situated at the anterior end of the body. If this view is correct, the question arises as to how far the posterior part of the nervous system of the Bilateralia can be regarded as derived from the primitive radial ring.

A circumoral nerve-ring, if longitudinally extended, might give rise to a pair of nerve-cords united in front and behind— exactly such a nervous system, in fact, as is present in

many Nemertines[1] (the Enopla and Pelagonemertes), in Peripatus,[2] and in primitive molluscan types (Chiton, Fissurella, &c.). From the lateral parts of this ring it is easy to derive the ventral cord of the Chætopoda and Arthropoda. It is especially deserving of notice, in connection with the nervous system of Nemertines and Peripatus, that the commissure connecting the two nerve-cords behind is placed on the *dorsal* side of the intestine. As is at once obvious, by referring to the diagram (fig. 18), this is the position this commissure ought, undoubtedly, to occupy if derived from part of a nerve-ring which originally followed more or less closely the ciliated edge of the body of the supposed radial ancestor.

The fact of this arrangement of the nervous system being found in so primitive a type as the Nemertines tends to establish the views for which I am arguing; the absence or imperfect development of the two longitudinal cords in Turbellarians may very probably be due to the posterior part of the nerve-ring having atrophied in this group.

It is by no means certain that this arrangement of the nervous system in some Mollusca and in Peripatus is primitive, though very probably it may be so.

In the larvæ of the Turbellaria the development of sense organs in the præoral region is very clear (fig. 9 B); but this is by no means so clear in the case of the true Pilidium. There is in Pilidium (fig. 19 A) a thickening of epiblast at the summit of the dorsal dome, which might seem, from the analogy of Mitraria, &c. (fig. 20), to correspond to the thickening of the præoral lobe, which gives rise to the supra-œsophageal ganglion; but, as a matter of fact, this part of the larva does not apparently enter into the formation of the young Nemertine (fig. 19). The peculiar metamorphosis which takes place in the development of the Nemertine out of the Pilidium,[3] may, perhaps, eventually supply an explanation of this fact; but at present it remains as a not yet explained difficulty.

The position of the flagellum in Pilidium, and of the supra-œsophageal ganglion in Mitraria, suggests a different view of the origin of the supra-œsophageal ganglion to that adopted above. The position of the ganglion in Mitraria corresponds

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[1] *Vide* Hubrecht, ' Zur Anat. and Phys. d. Nerven System d. Nemertinen Kon. Akad.,' Wiss., Amsterdam; and "Researches on the Nervous System of Nemertines," ' Journ. of Micr. Science,' 1880.

[2] *Vide* Self, "On some points in the Anat. of *Peripatus capensis*," ' Quart. Journ. of Micr. Science,' vol. xix, 1879.

[3] *Vide* ' A Treatise on Comparative Embryology,' vol. i, p. 169.

closely with that of the auditory organ in Ctenophora; and
it is not impossible that the two structures may have had a
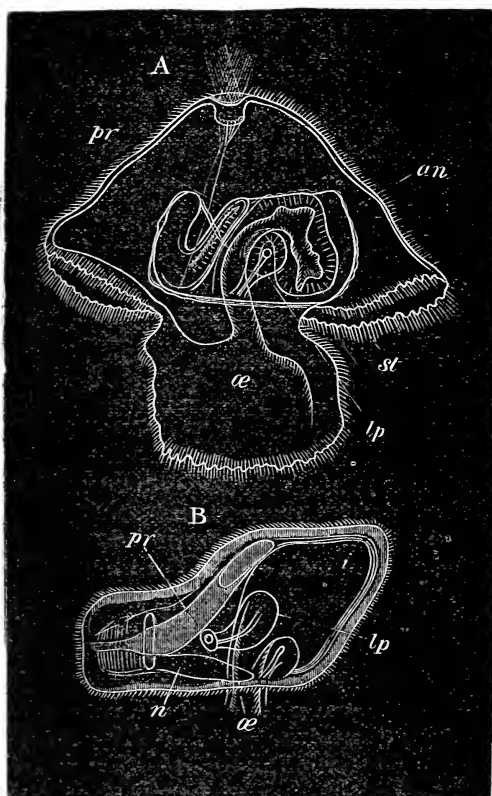
Fig. 19.—A. *Pilidium with an advanced Nemertine Worm.* B. *Ripe Embryo
of Nemertes in the position it occupies in Pilidium.* (Both after
Bütschli.) *œ.* œsophagus ; *st.* stomach ; *i.* intestine ; *pr.* proboscis ;
l.p. lateral pit ; *an.* amnion ; *n.* nervous system.

common origin. If this view is the correct one, we must
suppose that the apex of the aboral lobe has become the
centre of the præoral field of the Pilidium and Trochosphere
larval forms.[1] The whole of the questions concerning the
nervous system are still very obscure, and until further facts
are brought to light no definite conclusions can be arrived at.

The absence of sense organs on the præoral lobe of larval
Echinodermata, coupled with the structure of the nervous

[1] The independent development of the supra-œsophageal ganglion and
ventral nerve cord in Chætopoda (*vide* Kleinenberg, ' Development of *Lum-
bricus trapezoides*') suits this view very satisfactorily.

system of the adult, points to the conclusion that the adult Echinodermata *have retained*, and not, as is now usually held, secondarily acquired, their radial symmetry; and if this is admitted it follows that the obvious bilateral symmetry of Echinoderm larvæ is a secondary character.

The bilateral symmetry of many Cœlenterate larvæ (the larva of Æginopsis, of many Acraspeda, of Actinia, &c.), coupled with the fact that a bilateral symmetry is obviously advantageous to a free-swimming form, are quite sufficient to show that this supposition is by no means extravagant; while the presence of only two alimentary diverticula in Echinoderm larva is quite in accord with the presence of a single pair of perigastric chambers in the early larva of Actinia, though it must be admitted that the derivation of the water-vascular system from the left diverticulum is not easy to understand on this view.
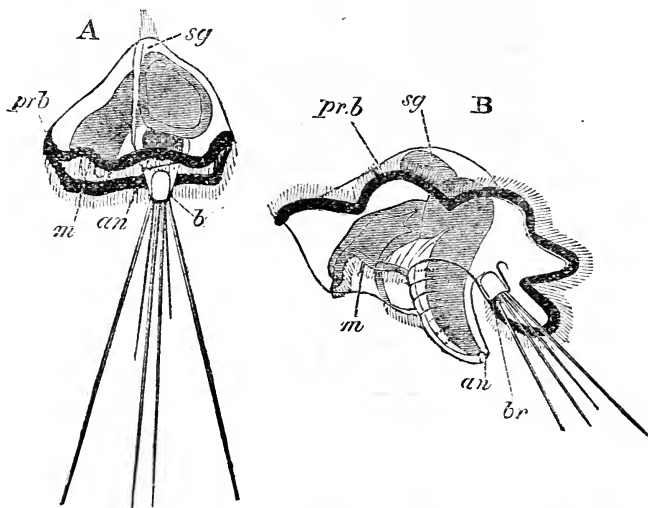
Fig. 20.—*Two Stages in the development of Mitraria.* (After Metschnikoff.) *m.* mouth; *an.* anus; *sg.* supra-œsophageal ganglion; *br.* and *b.* provisional bristles; *pr.b.* præoral ciliated band.

A difficulty in the above speculation is presented by the fact of the anus of the Echinodermata being the permanent blastopore, and arising prior to the mouth. If this fact has any special significance, it becomes difficult to regard the larva of Echinoderms and that of the other types as in any way related; but if the views already urged, in a previous essay on the germinal layers, as to the unimportance of the blastopore, are admitted, the fact of the anus coinciding with the blasto-

pore ceases to be a difficulty. As may be seen, by referring to fig. 18 c, the anus is placed on the dorsal side of the ciliated band. This position for the anus adapts itself to the view that the Echinoderm larva had originally a radial symmetry, with the anus placed at the aboral apex, and that the present terminal position of the anus arose with the elongation of the larva on the attainment of a bilateral symmetry.

It may be noticed that the obscure points connected with the absence of a body-cavity in most adult Platyelminthes, which have already been dealt with in my essay on the germinal layers, crops up again here ; and that it is necessary to assume either that alimentary diverticula, like those in the Echinodermata, were primitively present in the Platyelminthes, but have now disappeared from the ontogeny of this group, or that the alimentary diverticula have not become separated from the alimentary tract.

So far the conclusion has been reached that the architype of the six types of larvæ was a radiate form, and that amongst existing larvæ it is most nearly approached in general shape and in the form of the alimentary canal by the Pilidium group, and in certain other particulars by the Echinoderm larvæ.

The edge of the oral disc of the larval architype was probably armed with a ciliated ring, from which the ciliated ring of the Pilidium type and of the Echinodermata were most likely derived. The ciliated ring of the Pilidium varies greatly in its characters, and has not always the form of a complete ring. In Pilidium proper (fig. 19 A) it is a simple ring surrounding the edge of the oral disc. In Müller's larva of Thysanozoon (fig. 9 B) it is inclined at an axis to the oral disc, and might be called præoral, but such a term cannot be properly used in the absence of an anus.

The Echinoderm ring is oblique to the axis of the body, and, owing to the fact of its passing ventrally in front of the anus, must be called postoral.

The next point to be considered is the affinities of the other larval types to these two types.

The most important of all the larval types is the Trochosphere, and this type is undoubtedly more closely related to the Pilidium than to the Echinoderm larva. Mitraria amongst the Chætopods (fig. 20) retains, indeed, the form of Pilidium very closely, and mainly differs from a Pilidium in the possession of an anus and of provisional bristles ; the same may be said of Cyphonautes (fig. 21) amongst the Polyzoa.

The existence of these two forms appears to show that the

præoral ciliated ring of the Trochosphere may very probably
be derived directly from the circumoral ciliated ring of the
Pilidium; the other ciliated rings or patches of the Trocho-
sphere having a secondary origin.
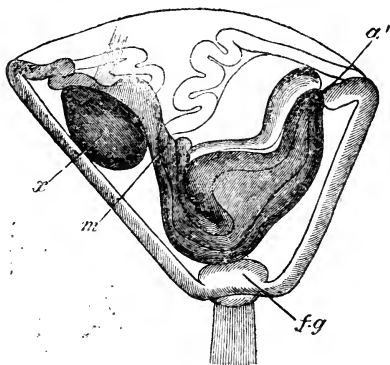
Fig. 21.—*Cyphonautes (Larva of Membranipora).* (After Hatschek). *m.*
mouth; *a.* anus; *f.g.* foot gland; *x.* problematical body (probably a
bud).

The larva of the Brachiopoda (fig. 6), in spite of its
peculiar character, is, in all probability, more closely related
to the Chætopod Trochosphere than to any other larval type.
The most conspicuous point of agreement between them is,
however, the possession in common of provisional setæ.

Echinoderm larvæ differ from the Trochosphere, not only
in the points already alluded to, but in the character of the
ciliated band. The Echinoderm band is longitudinal and
postoral. As just stated, there is reason to think that the
præoral band of the Trochosphere and the postoral band of
the Echinoderm larva are both derived from a ciliated ring
surrounding the oral disc of the prototype of these larvæ
(*vide* fig. 8). In the case of the Echinodermata the anus
must have been formed on the *dorsal side* of this ring, and
in the case of the Trochosphere on the *ventral side*; and so
the difference in position between the two rings was brought
about. Another view with reference to these rings has been
put forward by Gegenbaur and Lankester, to the effect that
the præoral ring is derived from the breaking up of the single
band of most Echinoderm larvæ into the two bands found
in Bipinnaria (*vide* fig. 10). There is no doubt a good deal
is to be said for this origin of the præoral ring, and it is
strengthened by the case of Tornaria; but the view adopted
above appears to me more probable.

Actinotrocha (fig. 17) undoubtedly agrees more closely with Echinoderm larvæ than with the Trochosphere. Its ciliated ring has the same character, and the growth along the line of the ciliated ring of a series of arms is very similar to what takes place in many Echinoderms. Its affinity with the Echinoderm larvæ is also shown in the absence of sense organs on the præoral lobe.

Tornaria (fig. 16) cannot be definitely united either with the Trochosphere or with the Echinoderm larval type. It has important characters in common with both of these groups, and the mixture of these characters renders it a very striking and well-defined larval form.

Phylogenetic conclusions.—The phylogenetic conclusions which follow from the above views remain to be dealt with. The fact that all the larvæ of the groups above the Cœlenterata can be reduced to a common type points to all the higher groups being descended from a single stem.

Considering that the larvæ of comparatively few groups have persisted, no conclusions as to affinities can be drawn from the absence of a larva ; and the presence in two groups of a common larval form may be taken as proving a common descent, but does not necessarily show any close affinity.

There is every reason to believe that the types with a Trochosphere larva, viz. the Rotifera, the Mollusca, the Chætopoda, the Gephyrea, and the Polyzoa, are descended from a common ancestral form, and it is also fairly certain there was a remote ancestor common to these forms and to the Platyelminthes. A general affinity of the Brachiopoda with these types very probable. All these types, together with any other types which can be proved to be related to them, are descended from a bilateral ancestor. The Echinodermata, on the other hand, are probably directly descended from a radial ancestor, and have more or less completely retained their radial symmetry. How far Actinotrocha[1] is related to the Echinoderm larvæ cannot be settled. Its characters may possibly be secondary, like those of the mesotrochal larvæ of Chætopods, or they may be due to its having branched off very early from the stock common to the whole of the forms above the Cœlenterata. The position of Tornaria is still more obscure. It is difficult, in the face of the peculiar water-vascular vesicle with a dorsal pore, to avoid the conclusion that it has some affinities with the Echinoderm larvæ. Such affinities would seem, on the lines of specula-

[1] It is very probable that Phoronis is in no way related to the other Gephyrea.

tion adopted in this essay, to prove that its affinities to the Trochosphere, striking as they are, are secondary and adaptive. From this conclusion, if justified, it would follow that the Echinodermata and Enteropneusta have a remote ancestor in common, but not that the two groups are in any other way related.

General conclusions and summary.—Starting from the demonstrated fact that the larval forms of a number of widely separated types above the Cœlenterata have certain characters in common, an attempt has been made(1) to determine the characters of the common prototype of all these larvæ,(2) and, the mutual relations of the larval forms in question. This attempt started with certain more or less plausible suggestions, the truth of which can only be tested by the coherence of the results which follow from them, and their capacity to explain all the facts.

The results arrived at may be summarised as follows :

1. The larval forms above the Cœlenterata may be divided into six groups enumerated on pages 391—394.

2. The prototype of all these groups was an organism something like a Medusa, with a radial symmetry. The mouth was placed in the centre of a flattened ventral surface. The aboral surface was dome-shaped. Round the edge of the oral surface was a ciliated ring, and probably a nervous ring provided with sense organs. The alimentary canal was prolonged into two or more diverticula, and there was no anus.

3. The bilaterally symmetrical types were derived from this larval form by the larva becoming oval, and the region in front of the mouth forming a præoral lobe, and that behind the mouth the trunk. The aboral dome became the dorsal surface.

On the establishment of a bilateral symmetry the anterior part of the nervous ring gave rise to the supra-œsophageal ganglia, and the optic organs connected with them. The body-cavity was developed from two of the primitive alimentary diverticula.

The usual view that radiate forms have become bilateral by the elongation of the aboral dome into the trunk is probably mistaken.

4. Pilidium is the larval form which most nearly reproduces the characters of the larval prototype in the course of its conversion into a bilateral form.

5. The Trochosphere is a completely differentiated bilateral form, in which an anus has become developed. The præoral ciliated ring of the Trochosphere is probably derived from

the ciliated ring of Pilidium, which is itself the original ring of the prototype of all these larval forms.

6. Echinoderm larvæ in the absence of a nerve ganglion or special organs of sense on the præoral lobe, and in the presence of alimentary diverticula, which give rise to the body cavity, retain some characters of the prototype larva which have been lost in Pilidium. The ciliated ring of Echinoderm larvæ is probably derived directly from that of the prototype by the formation of an anus on the dorsal side of the ring. The anus was very probably originally situated at the aboral apex.

Adult Echinoderms have probably retained the radial symmetry of the forms from which they are descended, and their nervous ring is probably directly derived from that of their ancestors. They have not, as is usually supposed, secondarily acquired their radial symmetry. The bilateral symmetry of the larva is, on this view, secondary, like that of so many Cœlenterate larvæ.

5. The points of similarity between Tornaria and (1) the Trochosphere and (2) the Echinoderm larvæ are probably adaptive in the one case or the other; and, while there is no difficulty in believing that those with the Trochosphere are adaptive, the presence of a water-vascular vesicle with a dorsal pore renders probable a real affinity with Echinoderm larvæ.

6. It is not possible in the present state of our knowledge to decide how far the resemblances between Actinotrocha and Echinoderm larvæ are adaptive or primary.

The majority of these conclusions are undoubtedly of a highly speculative character, but while they cannot be regarded as part of our stock of embryological knowledge, they may, nevertheless, serve to indicate an important line for continued embryological research. A thorough histological investigation of the larval forms dealt with in this essay will be likely to lead to valuable results.

On the CLASSIFICATION *of* CRYPTOGAMS.[1] By ALFRED W.
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    St. Thomas's Hospital.[2].

THE classification of Cryptogams proposed in the fourth
edition of Sachs's ' Lehrbuch der Botanik '—differing in
some important points from that in the third edition, from
which the English translation has been made—is an un-
doubted index of a considerable advance in our knowledge
of the relationships of the lower forms of flowerless plants.
That it will in some points be modified as our knowledge
further advances, its author would be the last to dispute ;
there are some other points to which I wish to call attention
in the present paper, in which that distinguished botanist
does not appear to me to have been so successful in pointing
out natural affinities.

The first group, the Thallophytes (including Characeæ), is
divided by Sachs into four classes of equal rank, the Proto-
phyta, Zygosporeæ, Oosporeæ, and Carposporeæ, abandoning,
as a primary classification, the time-honoured distinction of
Algæ and Fungi. Few will be disposed to dispute the
desirability of setting off the lowest forms from both the
Algæ and Fungi into a distinct class ; and the Protophyta
may with advantage be retained with the limits defined by
Sachs. But, beyond this, it is doubtful whether the pro-
posed change represents a nearer approach to a true phylo-
genetic scheme. The supporters of the new classification
point to that of Phanerogams as affording them powerful
support. They say, with truth, that genera destitute of
chlorophyll, like Orobanche, Balanophora, &c., are not
separated into a distinct class of primary rank. But the
cases are not parallel. Without going into the question of
whether these parasitic Phanerogams do contain chlorophyll
or not, it may be pointed out that they are mostly isolated
genera closely resembling well-known natural orders in all
important points of structure. Were flowering plants divi-
sible into two great series, one containing chlorophyll and
autonomous, the other destitute of chlorophyll and parasitic,
with very few points of resemblance or connecting links
between the two, it is at least doubtful whether this would

not be taken into account in the first line by systematists. The old division into Fungi and Algæ is so engrafted into popular conceptions, is so convenient to the student, and rests at the same time, with but few exceptions, on such well-marked physiological characters, that its abandonment requires the clearest evidence of the necessity for such a course, as being obviously in harmony with genetic affinities. But few will, we think, be bold enough to maintain that Spirogyra is more nearly allied to Mucor than it is to Œdogonium, or that the Florideæ display more affinity to the Basidiomycetes than they do to the Fucaceæ.

The present writer has already stated in detail ('Journal of Botany,' 1878, p. 202) his reasons for considering that it is impossible to place the Characeæ among Thallophytes, and still more among the Carposporeæ.

As regards minor points, I am disposed to consider the anomalous Myxomycetes as a low type of structure, scarcely raised above the Protophyta, and not exhibiting true sexual conjugation. The researches of Carpenter, Cohn, and others, clearly show that Volvox and its allies must be placed among the Oosporeæ rather than the Zygosporeæ. The Phæosporeæ should certainly be separated off as a distinct order from the Fucaceæ.

I propose, therefore, to divide Thallophytes into three primary classes:—PROTOPHYTA, FUNGI, and ALGÆ. The Protophyta are divisible into two sub-classes, *Protomycetes* and *Protophyceæ*. The Protomycetes consist of a single order, the Schizomycetes, of which Saccharomyces is regarded as an aberrant form. The Protophyceæ are composed of the Protococcaceæ (including Palmellaceæ and Scytonemeæ), Nostocaceæ, Oscillatorieæ, and Rivularieæ. The *Myxomycetes* are treated as a supplement to the Protophyta. The Fungi are made up of three sub-classes, employing in the main the same characters as Sachs; for reasons, however, which are stated in another paper presented to the Association, the syllable "sperm" is used instead of "spore" in their terminology. The first division, the *Zygomycetes* (or *Zygospermeæ achlorophyllaceæ*), is composed of the Mucorini only (including the Piptocephalidæ). The second, the *Oomycetes* (or *Oospermeæ achlorophyllaceæ*), comprises the Peronosporeæ and Saprolegnieæ (including the Chytridiaceæ). The third, the *Carpomycetes* (or *Carpospermeæ achlorophyllaceæ*), is made up of the Uredineæ, Ustilagineæ, Basidiomycetes, and Ascomycetes, the Lichenes being included in the last as a sub-order. The Algæ are arranged under three corresponding sub-classes. The *Zygophyceæ* (or

Zygospermeæ chlorophyllaceæ) is made up of the following orders :—Pandorineæ, Hydrodictyeæ, Confervaceæ (under which the Pithophoraceæ may possibly come), Ulotrichaceæ, Ulvaceæ, Botrydieæ, and Conjugatæ (the last comprising the Diatomaceæ, Desmidieæ, Zygnemaceæ, and Mesocarpeæ). The *Oophyceæ* (or *Oospermeæ chlorophyllaceæ*) include the Volvocineæ, Siphoneæ (with the nearly allied Dasycladeæ), Sphæropleaceæ, Oedogoniaceæ, Fucaceæ, and Phæosporeæ. The *Carpophyceæ* (or *Carpospermeæ chlorophyllaceæ*) is made up of the Coleochæteæ and Florideæ.

The CHARACEÆ will be restored to their old rank as a group of primary importance. The MUSCINEÆ are unchanged, comprising the *Hepaticæ* and *Musci* (including Sphagnaceæ).

In classifying the VASCULAR CRYPTOGAMS, I am compelled again to dissent from Sachs's most recent proposal. Abandoning the primary division into isosporous and heterosporous forms, he proposes in their place three classes, of which two are newly suggested—Equisetaceæ, Filicineæ, and Dichotomi; the two last including both isosporous and heterosporous forms, the Rhizocarpeæ being placed under Filicineæ, and the Selaginellaceæ under Dichotomi. The sexual differentiation of the spores into those which produce female prothallia and those which produce antherozoids, seems to me to lie so much deeper than any characteristics derived merely from vegetative characters, and to mark so distinctly a step upwards in the scale towards flowering plants, that I have no hesitation in restoring the primary division of vascular cryptogams into *Isosporia* and *Heterosporia*. As a minor point, it hardly seems logical to separate the Ophioglossaceæ as a distinct order from Filices, if the Marattiaceæ are to remain associated with them. The Isosporia will then consist of the Filices (including Ophioglossaceæ), Lycopodiaceæ, and Equisetaceæ. The Heterosporia will comprise the Rhizocarpeæ and Selaginellaceæ.

In describing the heterosporous vascular cryptogams it is almost universal to speak of the spores which give rise to the female prothallium and those which give birth to antherozoids as "macrospores" and "microspores" respectively. The first of these terms is doubly objectionable; firstly, etymologically, the correct meaning of μακρός being not large, but long; and, secondly, from the close similarity in sound of the two terms, an inconvenience, especially in oral instruction, which every teacher must have experienced. Seeing that the correct and far preferable terms *megaspore* and *microspore* are used by Berkeley, Areschoug, and others,

it is difficult to understand how "macrospore" can ever have got into general use.[1]   But those terms are always to be preferred which convey to the student's mind important rather than unimportant facts; and since, in the present case, the sexual differentiation is far more important than the difference in size, I propose that the two kinds of spores shall in future be known as *gynospores* and *androspores*, the receptacles in which they are produced as *gynosporangia* and *androsporangia* respectively.

Appended is a table of the proposed classification.

Proposed Classification of Cryptogams.

A. PROTOPHYTA.
  a. Protomycetes (Protophyta achlorophyllacea).
    Schizomycetes.
     Saccharomyces.
  b. Protophyceæ (Protophyta chlorophyllacea).
    Protococcaceæ.
     Palmellaceæ.
     Scytonemeæ.
    Nostocaceæ.
    Oscillatorieæ.
    Rivularieæ.
 Supplement to Protophyta.   *Myxomycetes.*

B. FUNGI.
  a. Zygomycetes (Zygospermeæ achlorophyllaceæ).
    Mucorini.
     Piptocephalidæ.
  b. Oomycetes (Oospermeæ achlorophyllaceæ).
    Peronosporeæ.
    Saprolegnieæ.
     Chytridiaceæ.
  c. Carpomycetes (Carpospermeæ achlorophyllaceæ).
    Uredineæ.
    Ustilagineæ.
    Basidiomycetes.
    Ascomycetes.
     Lichenes.

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[1] The equally objectionable term "macrolepidoptera" seems to have crept into entomological terminology.

C. Algæ.
    a. Zygophyceæ (Zygospermeæ chlorophyllaceæ).
        Pandorineæ.
        Hydrodictyeæ.
        Confervaceæ.
          ? Pithophoraceæ.
        Ulotrichaceæ.
        Ulvaceæ.
        Botrydieæ.
        Conjugatæ.
          Diatomaceæ.
          Desmidieæ.
          Zygnemaceæ.
          Mesocarpeæ.
    b. Oophyceæ (Oospermeæ chlorophyllaceæ).
        Volvocineæ.
        Siphoneæ.
          ? Dasycladeæ.
        Sphaeropleaceæ.
        Oedogoniaceæ.
        Fucaceæ.
        Phæosporeæ.
    c. Carpophyceæ (Carpospermeæ chlorophyllaceæ)
        Coleochaeteæ.
        Florideæ.

D. Characeæ.

E. Muscineæ.
        Hepaticæ.
        Musci.
          Sphagnaceæ.

F. Cryptogamia Vascularia.
    a. Isosporia.
        Filices.
          Ophioglossaceæ.
        Lycopodiaceæ.
        Equisetaceæ.
    b. Heterosporia.
        Rhizocarpeæ.
        Selaginellaceæ.

A Reformed System *of* Terminology *of the* Reproduc-
tive Organs *of the* Cryptogamia. By Alfred W.
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We have been led to the following attempt at obtaining a
symmetrical system of terminology among Cryptogams by the
undoubted fact that the anomalies at present existing greatly
retard the prosecution of the study of this most interesting
group of plants. A more striking instance of the want of
accuracy in the use of the commonest terms could not be
afforded than by observing the different meanings, often
quite irreconcilable with one another, applied by the most
approved writers to the term "spore." Thus, Le Maout and
Decaisne (Hooker's edition of 'Descriptive and Analytical
Botany,' p. 14) and Professor Asa Gray ('Botanical Text-
book,' sixth edition, p. 434) speak of spores as "the ana-
logues of seeds;" Sachs ('Text-book,' English edition, p. 203,
but modified in the most recent German edition) defines
them as "asexual reproductive cells." In direct opposition
to each and both of these definitions, Berkeley ('Micrographic
Dictionary,' third edition, p. 327) describes the unfertilised
ova or oospheres of Fucus as spores; and Huxley and Mar-
tin ('Elementary Biology,' p. 45) call the archegonium of
Characeæ a spore-fruit or sporangium. A still more sin-
gular example occurs in the most recent text-book of botany
introduced to English readers (Vines's edition of 'Prantl's
Text-book" where we read on one page (p. 97) "Reproduc-
tion of Cryptogams is effected asexually by cells termed
gonidia, conidia, or spores;" and on another page (p. 115),
" Fungi are reproduced sexually by means of spores."

The object kept before us in this attempt at reform has
been to arrive at a system which shall be symmetrical and
in accordance with the present state of knowledge, and which
shall, at the same time, interfere as little as possible with
existing terms. In preparing such a system it has been
impossible to avoid introducing several new terms, but these
are associated with one another on an etymological plan
which will not burden the memory of the student, while the
total number of terms in cryptogamy will be greatly reduced

[1] Read at the Swansea Meeting of the British Association, Aug. 26th,
1880.

by the abandonment of a large number that are absolutely useless.

So far as we know, the first systematic attempt at placing cryptogamic terminology on a more satisfactory footing was made by Sachs in the fourth edition of his ' Lehrbuch ' (not yet published in English), which is deserving of commendation, though it does not appear to us altogether successful. Sachs proposes (p. 243) to define a " spore " as a reproductive cell produced " *directly or indirectly* by an act of fertilisation," reserving the term " gonidium " for those reproductive cells which are produced without any previous act of impregnation. As far as Vascular Cryptogams and Muscineæ are concerned, Sachs's proposal involves no change, since he regards the spores, which are a part of the nonsexual generation, as the indirect result of the act of fertilisation which closes the sexual generation. In the Basidiomycetes he is able to retain the term spore for the familiar bodies commonly so called, only by the assumption —for at present it is nothing more—that the structure of which the hymenium forms a part is the result of a yet undiscovered process of fertilisation on the mycelium. In the lower fungi the changes involved in the proposal are considerable. Thus the " spores " of Penicillium, similar as they are to those of Agaricus, can no longer be called spores, because they do not result from an act of fertilisation, but become " conidia " or " gonidia;" and for the same reason the familiar " zoospores " of the lower Thallophytes become " zoogonidia." It is obvious that one practical defect of this suggestion is that it may necessitate a perpetual change of terminology as our knowledge advances. Every fresh extension of the domain of sexual fecundation— and it is probable that many such will take place—will involve the removal of a fresh series of reproductive cells from the category of gonidia to that of spores, even though they may not be the immediate result of an act of fertilisation. Again, if the spores of Ferns and Mosses are the indirect result of impregnation, it is difficult to say why the term should not ultimately include all reproductive bodies whatever, except the spores of the " apogamous ferns " with which Farlow and De Bary have recently made us acquainted, and of other similar abnormal productions, which are certainly not the result of impregnation, direct or indirect.

It seems a sounder principle—and is certainly more convenient to the student—to base a system of terminology on facts which can be confirmed by actual observation, rather

than on unproved hypotheses. We propose, therefore, as the basis of our terminology, to restore the term *spore* to what has been in the main hitherto its ordinary signification, and to restrict its use to *any cell produced by ordinary processes of vegetation, and not directly by a union of sexual elements, which becomes detached for the purpose of direct vegetative reproduction*. The spore may be the result of ordinary cell division or of free cell formation. In certain cases (*zoospore*) its first stage is that of a naked primordial mass of protoplasm. In rare instances it is multicellular, breaking up into a number of cells (*polyspore*, composed of *merispores*, or breaking up into *sporidia*).

The simple term *spore* will, for the sake of convenience, be retained in Muscineæ and Vascular Cryptogams; but in the Thallophytes it will always be used in the form of one of those compounds to which it so readily lends itself, expressive of the special character of the organ in the class in question. Thus, in the Protophyta and Mucorini, we have *chlamydospores ;* in the Myxomycetes, *sporangiospores ;* in the Peronosporeæ, *conidiospores ;* in the Saprolegnieæ, Oophyceæ, and some Zygophyceæ, *zoospores ;* in the Uredineæ, *teleutospores, æcidiospores, uredospores,* and *sporidia ;* in the Basidiomycetes, *basidiospores ;* in the Ascomycetes (including Lichenes), *conidiospores, stylospores, ascospores, polyspores,* and *merispores ;* in the Hydrodictyeæ, *megaspores ;* in the Desmidieæ, *auxospores ;* in the Volvocineæ and Mesocarpeæ, *parthenospores ;* in the Siphoneæ and Botrydieæ, *hypnospores ;* in the Oedogoniaceæ, *androspores ;* in the Florideæ, *tetraspores* and *octospores*. The cell in which the spores are formed will, in all cases, be called a *sporangium*. It is obvious that, if greater precision is desired, this term might be compounded in the same way as spore ; but the words thus formed would be needlessly cumbrous for ordinary use.

The male organs of fecundation are so uniform in their structure throughout cryptogams that very little complication has found its way into their terminology. The cell or more complicated structure in which the male element is formed is uniformly known among Cormophytes as well as Thallophytes as an *antheridium ;* the fecundating bodies are almost invariably naked masses of protoplasm, provided with vibratile cilia, endowed with apparently spontaneous motion, and bearing the appropriate name of *antherozoids* or " spermatozoids." The former of these is preferable for two reasons ; from its etymological connection with antheridium, and because the use of terms compounded from

" sperm " should, for reasons to be detailed presently, be avoided for male organs. In only two important groups, Florideæ and Lichenes, are the fecundating bodies destitute of vibratile cilia and of spontaneous motion ; in the former case they are still usually termed antherozoids ; in the latter " spermatia," and their receptacles " spermogonia." In order to mark the difference in structure from true antherozoids, it is proposed to designate these motionless bodies in both cases *pollinoids ;* the term spermogonium is altogether unnecessary, the organ being a true antheridium.

A satisfactory terminology of the female reproductive organs presents greater difficulties, from the much greater variety of structure, and the larger number of terms already in use. The limits we have placed to the use of the term " spore " and its compounds require the abandonment of " oospore " for the fertilised ovum or oosphere in its encysted state (enclosed in a cell-wall), anterior to its segmentation into the embryo ; and this is the most important change involved in the new system.

In devising a term which shall include all those bodies which are the immediate result of impregnation, it was necessary to take two points specially into account: Firstly, the term must be capable of defence on etymological grounds ; and secondly, it must, like spore, be capable of ready combination. After much consideration we have decided on proposing the syllable *sperm.* No doubt the objection will present itself that the Greek σπέρμα, like the Latin " semen," and the English " seed " (as used by old writers), while originally meaning the ultimate product of fertilisation, came afterwards to signify the male factor in impregnation ; and hence, in zoology, terms derived from these roots are used for the male fertilising bodies. But the objection applies to a much smaller extent to phyto-terminology. Some terms compounded from " sperm," as gymnosperm, angiosperm, &c., are already familiarly in use in a sense similar to that we would indicate ; while of those used in the reversed sense, " sperm-cell," for antherozoid or pollen-grain, has never come into general use in this country ; " spermatozoid " is easily replaced by antherozoid ; " spermogonium " is simply a peculiar form of antheridium, and " spermatium " has already been referred to. Accepting this term as the least open to objection of any that could be proposed, it will be found to supply the basis of a symmetrical system of terminology, which will go far to redeem the confusion that at present meets the student at the outset of his researches. For the unfertilised female protoplasmic mass the term

oosphere is already in general use ; and, though not all that could be desired, it is proposed to retain it, and to establish from it a corresponding series of terms ending in *sphere.* The entire female organ before fertilisation, whether unicellular or multicellular, is designated by a set of terms ending in *gonium*, again following existing analogy.

As will be seen from another paper presented to the Association, we propose, in our forthcoming " Handbook of Cryptogamic Botany," to make the primary division of the Thallophytes into the three great classes of Protophyta, Fungi, and Algæ, each of the two last being again divided into three parallel series of Zygospermeæ, Oospermeæ, and Carpospermeæ, corresponding nearly to Sachs's Zygosporeæ, Oosporeæ, and Carposporeæ. In the Zygomycetes and Zygophyceæ, the conjugated *zygospheres*, or contents of the *zygogonia*, constitute a *zygosperm ;* in the Oomycetes and Oophyceæ, the fertilised *oosphere,* or contents of the *oogonium,* is an *oosperm ;* in the Carpophyceæ, the fertilised *carposphere,* or contents of the *carpogonium,* constitutes a *carposperm.* In this last class the process is complicated, being effected by means of a special female organ, which, to keep up the etymological analogy, may be called a *trichogonium* rather than a " trichogyne." The ultimate result of impregnation is the production of a mass of tissue, known as the *cystocarp* (or " sporocarp "), within which are produced the germinating bodies, which must be designated *carpospores,* since they are not the direct result of fertilisation, but which must be carefully distinguished from the so-called " carpospore "—properly an archesperm—in the Characeæ. Any of these bodies which remains in a dormant condition for a time before germinating will be a *hypnosperm.*

In the Zygospermeæ it is no doubt the case that the two conjugating bodies are really a zygosphere and an antherozoid (or pollinoid) ; but, as they are at present absolutely indistinguishable, it seems best to sacrifice theory, and call them both zygospheres.

It may be mentioned, by way of completing the analogy, that a precisely analogous set of terms will be proposed for the Cormophytes (Characeæ, Muscineæ, and Vascular Cryptogams), throughout which the fertilised *archesphere,* or contents of the *archegonium,* is called an *archesperm.* The latter term may be objected to as nearly identical with " archisperm," used by Strasburger and others as a synonym for gymnosperm ; but the term has not come into general use, and no confusion between the two

seems possible. In the proposed system zygosperm will replace Strasburger's "zygote," and the "gametes" of the same writer will be zygospheres; while his "zoogametes" or "planogametes" must enjoy the somewhat cumbrous name of "zoozygospheres," the prefix "zoo" or suffix "zoid" being always used to denote an apparently spontaneous power of motion.

In the Basidiomycetes, Ascomycetes, and some other classes of Cryptogams, the entire non-sexual generation which bears the spores is frequently called the "receptacle," a term which is for several reasons objectionable. In the first place it has no meaning etymologically in this connection; and secondly, it suggests a false analogy with the receptacle in flowering plants, a structure which supports the sexual reproductive organs. A convenient expression for that portion of the non-sexual generation which bears the spores is the *fructification*, corresponding to the German equivalent "Fruchtkörper," and already popularly used in this sense in the case of mosses, as well as for the sori of ferns. Except, as far as is yet known, in the case of the Basidiomycetes, the fructification is always the result of impregnation; and, according to Reess, Eidam, and others, the same is the case also in that class of fungi.

To illustrate the simplification effected in cryptogamic terminology by the proposed reform, two comparative tables are appended; and the following list is given, which enumerates some of the terms in more frequent use by various writers as respects Thallophytes and Characeæ, all of which are disused in the proposed system.

Non-sexual organs.—Gemma, conidium, gonidium, endo-gonidium, zoogonidium, macrozoospore, acrospore, ectospore, swarmspore, pycnidium, perithecium, apothecium, theca, receptacle.

Male sexual organs.—Spermatozoid, spermatium, spermo-gonium, globule.

Female sexual organs.—Oospore, zygospore, zygogonidium, zygozoospore, zygote, gamete, planogamete, aplanogamete, ascogonium, nucule, sporocarp, ceramidium, coccidium, favella, favellidium, sphaerospore.

Modes of Fertilization in Cryptogams.

Conjugating Bodies.

ZYGOSPERMEÆ. Zygogonia, containing Zygospheres.

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(fertilised) Zygosperm.

	Male Organ.	*Female Organ.*
OOSPERMEÆ.	Antheridium, containing	Oogonium, containing
	Antherozoids or ⎱ =	Oosphere.
	Pollinoids. ⎰	

‖

(fertilised) Oosperm.

CARPOSPERMEÆ.	Antheridium, containing	Carpogonium, containing
	Antherozoids or ⎱ =	Carposphere.
	Pollinoids. ⎰	

‖

(fertilised) Carposperm.

CORMOPHYTA.	Antheridium, containing	Archegonium, containing
	Antherozoids =	Archesphere.

‖

(fertilised) Archesperm.

Productive Organs of Thallophytes.

	Female.	*Non-sexual.*
PROTOPHYTA.		Chlamydospore. Sporangium.
MYXOMYCETES.		Zoospore. Sporangiospore.
MUCORINI.	Zygogonium. Zygosphere. Zygosperm.	Chlamydospore. Sporangiospore.
PERONOSPOREÆ.	⎧ Oogonium. Oosphere.	Sporangium. Conidiospore. Zoospore.
SAPROLEGNIEÆ.	⎨ Oosperm.	Zoospore.
UREDINEÆ.	⎧ Carpogonium. Carposphere.	Teleutospore. Aecidiospore. Uredospore. Sporidium.
USTILAGINEÆ.	⎨ Carposperm.	Teleutospore. Sporidium.
BASIDIOMYCETES.		Basidiospore. Sterigma. Basidium.

ASCOMYCETES, including LICHENES.	Trichogonium. Pollinodium.	Conidiospore. Stylospore. Ascospore. Polyspore. Merispore.
ZYGOPHYCEÆ.	Zygogonium. Zygosphere. Zoozygosphere. Zygosperm. Hypnosperm (Hydrodictyeæ zygnemaeæ).	Zoospore. Megazoospore (Hydrodictyeæ). Auxospore (Diatomaceæ). Hypnosporangium ⎫ Hypnospore ⎬ Botrydieæ. Parthenospore (Mesocarpeæ).
OOPHYCEÆ.	Oogonium. Oosphere. Oosperm. Conceptacle. Hypnosperm.	Zoospore. Parthenospore (Volvocineæ). Androspore (Oedogoniaceæ). Hypnospore (Siphoneæ).
CARPOPHYCEÆ.	Carpogonium. Carposphere. Carposperm. Trichogonium. Trichophore. Cystocarp.	Zoosporangium. Tetraspore. Octospore. Carpospore.

On the LAMINAR TISSUE *of Amphioxus.* By Professor POUCHET. (With Plate XXIX.)

THE chief results in this paper were communicated to the Société de Biologie at their meeting on May 1st, 1880.[1]

I. I was lately fortunate in obtaining large numbers of Amphioxus at Concarneau, and I used them in endeavouring to clear up a special point in their histology, which seems to have been only very imperfectly touched upon by the numerous authors who have studied this animal. I intend to speak of the system of canals and cavities, which from a point of view, which is perfectly legitimate, are comparable to fin rays. Both of these systems are differentiations of what may be termed "laminar tissue."

In 1849 M. de Quatrefages applied the term "singular" to the laminar tissue of Amphioxus. Since that time the system of spaces and canals which have been observed have not been the object of special study. The fact has been recognised that they do not communicate with the blood capillaries, but their histological structure has remained obscure. M. Reichert is the only author who has touched upon it, and he seems to have guessed at, rather than proved, their analogy with the constant elements of the laminar tissue. He expresses himself thus, "Da das bindegewebige Stroma uns als pellucide Grundsubstanz angesehen werden Kann und die dazu gehörigen Bindesubstanzkörper fehlen, so wäre es möglich dass das in Rede stehende netzförmige Gebilde den zellenhalftigen Theil des bindegewebigen Stromas darstellt, unter dessen Vermittelung die in grosser Menge verbreitete, ganz hyaline Grundsubstanz gebildet werde."[2]

In 1873 Stieda[3] contents himself with quoting these words of Reichert, which in fact contain the true state of the case.

Before Reichert's paper was published, Owsjannikow[4] described, in 1868, in the region of the tactile corpuscles, that is to say, round the mouth, connective-tissue cells, "gross, länglich, zuweilen sternförmig," and compared them to the cells of the cornea, an error of observation as will be seen further on.

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[1] 'See 'Gazette Médicale' for May 22, 1880.

[2] Reichert, 'Arch. f. Anat.,' 1870.

[3] "Studien über den Amphioxus lanceolatus," 'Mém. de l'Académie de St. Petersberg,' viie Série, Tome xix, No. 7, 1873.

[4] "Ueber das centrale Nervensystem des Amphioxus lanceolatus," in Bullet. de l'Acad. de St. Petersbourg,' T. xii, 1868, p. 299.

P. Langerhans, in 1873, mentions[1] the same cells; ne describes them as very much branched (reich verästelt), and compares them, as does Owsjannikow, to the cells of the cornea, and corroborates the description of Stieda, although this is limited to the quotation from Reichert, which rather goes against the existence of such cells.

Quite recently Ant. Schneider[1] admits the truth of Reichert's ideas, and at the same time describes (pp. 1 and 28) and figures in the laminar tissue of Amphioxus stellate fibro-plastic cells, inserted by their prolongations on the walls of the true capillaries around the mouth.

These quotations suffice to show the confusion and incompleteness of our knowledge of the laminar tissue of Amphioxus, and of the formed elements which it presents. As we shall presently show, even if it is possible to find isolated cells having the ordinary appearance of stellate fibro-plastic cells, it is quite exceptional at least in the adult. In our preparations we have found one or two in the thinnest region of the caudal lophioderm, quite at the extremity; with this exception we have not come across them. It is in fact a very decided character of the laminar tissue of Amphioxus that the formed elements analogous to the fibro-plastic cells never appear in an isolated state, and that they are always united again into scattered groups or in longer or shorter rows which are more or less regular, and anastomose to a greater or less extent, forming cords which are sometimes solid and sometimes hollow for a certain distance, or even form large spaces.

It is these cords, which are partly solid, partly hollowed, and generally slightly elongated, which have been described as "systems of canals." The supposed branched cells of Owsjannikow are only the enlargements of this very irregular network in the neighbourhood of the mouth.

II. As these cordons and spaces formed by the coalescence of the cells of the connective tissue are situated to a large extent in the layer of laminar tissue which surrounds the body of the animal, we shall first of all describe this,[3] adding, however, but little to the descriptions of Stieda.

Below the epidermis you get, passing from without inwards—

[1] "Zur Anatomie des Amphioxus lanceolatus," in 'Arch. f. Mik. Anat.,' 1873, p. 301.

[2] 'Beiträge zur vergleichenden Anatomie,' 4to, Berlin, 1879.

[3] Compare with this the structure of the skin in fishes. See Pouchet, "Du développement des poissons osseux," 'Journal de l'Anatomie,' May-June, 1879, pp. 288-289.

1. The dermis.   2. The subdermic layer.   3. The subcutaneous aponeurosis.

1. *The dermis*[1] is formed, as in fishes and Amphibia, of a thin layer which appears in section to be homogeneous, and which in Amphioxus presents upon its external surface a double rectangular striation, with the *crosses* corresponding to the nerve terminations which Langerhans[2] has so well figured.

2. *Subdermic layer.*—The subdermic layer[3] is characterised in Amphioxus by the presence of a large amount of structureless substance.   This latter is as usual absolutely hyaline, and keeps this character even after the action of osmic acid in saturated solution.[4]   It is traversed by fibres having the character of laminar fibres, stretching from the deep face of the dermis to the subcutaneous (cf. fig. 3A). These fibres are absolutely devoid of any nucleus and of any division into cells, a fact not without interest as bearing upon the much debated history of the genesis of the laminar fibres. We have pointed out in the following terms[5] the existence of similar fibres in the lophioderm of larval Axolotls measuring 60 to 80 mm. in length : " In sections cut 'à la planchette,' after treatment with osmic acid, the amorphous matter is seen to be traversed by vessels and nerves, and to enclose some fibro-plastic cells with regular spaces between them, invisible ; but extremely fine laminar fibres are also seen.   These run from the skin of one side to that of the other, in the same manner as the strings in an instrument in a very beautiful manner, all parallel to one another. These fibres seem, therefore, to have no relation to the fibro-plastic cells."

It was always difficult in Axolotl to pronounce an opinion on the independence of the fibres and cells on account of the presence of the latter.   Whilst in Amphioxus there are no fibro-plastic cells furnished with any prolongations from which laminar fibres might be derived.   These are also larger than in Axolotl, and are more deeply stained by carmine.   No migratory corpuscles are found in the hyaline structureless portion of the subdermic tissue of Amphioxus.

3. *Subcutaneous aponeurosis.*—This membrane appears in

[1] " Cutis oder Lederhaut," Stieda.
[2] Loc. cit., fig. 13.
[3] " Das Unterhautgewebe " of Stieda.
[4] For the action of this reagent see Pouchet, " De l'emploi des solutions concentrées d'acide osmique," ' Journal de l'Anat.,' Sept.-Oct., 1876.
[5] G. Pouchet et Tourneux, ' Précis d'histologie humaine et d'histogenie,' 1878, p. 102.

section to be finely striated as in fishes. A lamellar struc-
ture must be attributed to it, and elongated nuclei appear in
certain places. It belongs to a system of fibrous tissue widely
distributed in the body of the animal ; this is quite distinct
from the laminar system properly so called, while in its
physico-chemical characters it approaches the dermis. To
this system belong the aponeuroses which separate the muscles
and envelope the medulla, &c. The skin, the subdermic apo-
neurosis, and the fibrous partitions of which we have been
speaking, when treated with osmic acid and picrocarmine,
take a splendid red staining, while the structureless sub-
stance of the subdermic layer and other varieties of struc-
tureless substance which are sometimes found in the body of
Amphioxus, remain uncoloured or become slightly yellow,
but do not take the carmine.

III. We have said that the special character of the formed
elements of the laminar tissue of Amphioxus was that they
are never present in an isolated state nor as cells simply
united, like the cells in the cornea of Amphibia, by fine
prolongations. We shall first describe these elements and
the network which they form in the "lophioderm"[1] of the
caudal fin of Amphioxus, and afterwards their modifications
in other parts of the body of the animal.

The caudal " lophioderm " presents a network with regular
elongated meshes, which had already attracted the attention
of M. de Quatrefages, and which at once calls to mind the
figure which he gave at a time when the technical methods
necessary for such a study were not yet in vogue.

This network is arranged in one plane in the centre of
the lophioderme, which is very thin in this region. The
meshes of the network are elongated in the neighbourhood
of the anus, and measure 60 μ to 80 μ in length, by 10 μ
in breadth (see fig. 1). The substance of the network is
formed of rods, which measure generally 8 to 10 μ; they
may, however, be narrower or broader than this. They are,
in the region of which we are speaking, flattened out ; they
are solid. At the intersections of the network there are
generally 6 to 10 larger or smaller nuclei. These are oval
or almost spherical, irregularly placed, and separated by a
distance which is generally less than their diameter. Some-
times several are placed in a row in one of the rods. They
are then generally more or less obliquely placed. The rods
after the action of osmic acid stain slightly with picrocar-

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[1] *Lophioderm* is the termed used by M. Pouchet for the dermal fold
which forms the substance of the median fin in Amphioxus and in Am-
phibia.—ED.

mine. They represent the bodies of the cells united one with another, or rather, all fused together.[1]

The only tissue which approaches in histological structure that which we are about to describe, is the tissue of the cornea in the eyes of bony fish. We have elsewhere described this tissue thus :—" All the corneal cells appear fused together, so that it is impossible to distinguish their outline, the protoplasmic bodies of the cells forming a sort of membrane, with nuclei scattered here and there. The meshes of this network are small and separated by extremely broad rods, which forms a true lamellar system mixed up with the fundamental lamellar system of the organ, &c."[2]

In the lophioderm of Amphioxus the meshes of the network are broader than the rods, and these latter are disposed in the same plane ; but the two tissues, nevertheless, present an analogy which enables one to place them side by side among the large number of varieties which the laminar tissue of vertebrates presents for our study.

Fig. 6 shows that in posterior region of the notochord the nuclei are very abundant ; they are very small, and answer very well to the description and figure given by Stieda. On the other hand, they are scattered throughout the substance of this organ, which does not agree with the opinion of Kossman (see ' Verhandlungen der Phys. Med. Ges.,' Wurzburg, 1874).

In the deep layer under the dermis in the neighbourhood of the mouth, the network of cells of the laminar tissue has not the same appearance. It presents large irregular meshes, with a slightly sinuous contour. The nuclei are no longer accumulated at the junctions of the network. The rods are almost cylindrical, and are swollen here and there.[3] These swellings present a larger or smaller hollow in the centre, bounded on all sides by the protoplasm and the nuclei, which are arranged in a single row. In optical section these hollows appear to be simply fissures between the layers of cellular substance.

Sometimes these enlarge and spread to a greater or less extent, but they always remain very irregular. This is the

[1] It sometimes happens in certain preparations that at the intersections of the network there is a sort of cleavage, so that the nuclei appear to be each surrounded by a protoplasmic body with a polygonal outline.

[2] G. Pouchet et Tourneux, loc. cit., p. 611, et fig. 168.

[3] These swellings, scattered on the thinnest rods of the network, have no doubt given rise to the error of Owsjannikow, who thought he had seen the cellular bodies anastomising by their prolongations.

"system of canals"; it is very different from a capillary
system properly so called, and its true nature has
been very variously interpreted by different authors. In
the neighbourhood of the notochord these cavities appear
to enlarge and spread out. Fig. 3 B shows one of these
cavities, pyriform in shape, the extremity being narrowed
and running into a rod, which binds it to the network.
Thus, this sort of hollow swellings forms a transition to
other cavities which are quite separate from the network of
cells, and which we are about to describe.

IV. The upper surface of the anterior extremity of the
notochord presents isolated groups of connective-tissue
cells, which completely envelope the cavities which they
bound, being arranged in a single row, as in the swellings
of the network in the neighbourhood of the mouth.

These groups of cells, always clearly bounded on the
outside, are generally oval in form, or rather, polyhedral,
where they are crowded together. The nuclei presents
slight prominences in their interior, and, in certain
individuals the cells contain various pigment granules of
a reddish colour.[1]

The interior of the cavity is filled with a liquid, which is
unacted on by osmic acid, and does not stain with most
reagents. It appears to be watery. The further we recede
from the extremity of the cord the larger do these laminar
cavities become (see fig. 2). They become also slightly
modified, and finally constitute the organs described under
the very bad name "fin rays" (see figs. 4 and 5). The
structure of these organs is extremely simple. They
are cellular spaces, larger than the others, almost cubical
in form, one wall of which, that which looks towards
the notochord, is invaginated into the interior of the cavity
itself, so as to fill up two thirds or three fourths of it. We
call the invaginated portion the *papilla*. It is made of a
structureless substance, presenting special characters which
clearly distinguish it from that of the sub-dermic layer.

Stieda describes these parts very incompletely. He states,
it is true, the analogy between these cavities and the system
of canals, and describes their cellular covering, but the in-
vaginated portion which Müller had already mentioned he
has overlooked. Müller states that in the interior of these
cavities there exists " eine durchsichtige Flüssigkeit und eine
consistentere, aber weiche Masse." The cavities are bounded
at the sides and below by the subcutaneous aponeurosis,

[1] Some specimens of Amphioxus present a reddish tint, others a
greenish.

and are separated from one another by transverse partitions (see fig. 5), which are very thin.[1]   The whole cavity, the papilla as well as the walls, is lined with a layer of cells, which closely resemble those lining the other spaces ; there is no appreciable difference between the cells on the papilla and those on the walls.   The contained liquid seems to be the same as that in the other spaces.

In the living animal the substance of the papilla can only be distinguished from the ambient liquid by the optical effects due to their differing density.   In a specimen which has been preserved in alcohol the papilla had almost disappeared. Certain reagents have the same action, whilst others appear to swell them up.   If the specimens are put into alcohol, even after the action of osmic acid, the papilla are altered in shape and drawn in.   Their surface presents depressions, which gives them a jagged appearance in section.[2]

The use of reagents brings to view minute cavities inside the papilla, more or less irregular, and about the size of one of the nuclei.   Suitable methods show that the amorphic substance of the papillæ contains nothing but some very fine and scattered fibres.   Transverse sections (fig. 4) show very well the relations of the papillæ.   There is a fibrous lamina occupying the middle line of the body, which, bifurcating at the level of the upper edge of the two lateral muscular masses, joins on each side the subcutaneous aponeurosis.   The base of the papilla rests in the groove formed by this bifurcation, and is clearly marked out by its physico-chemical characters.   The cavity which it contains, is itself bounded laterally and below by the subcutaneous aponeurosis.

The papilla is traversed towards its base by a layer of very fine fibres (fig. 4, c.) stretched between the two fibrous laminæ.   In front and behind (fig. 5, c.) this sheet of fibres becomes raised up before being inserted into the partitions which separate the cavities.   On the upper face of the layer a very loose and irregular network of extremely fine laminar fibres rise up and spread out in the centre of the papilla.   None of these are directed downwards.   When treated with osmic acid and then saturated with picro-carmine, the substance of the papilla assumes a slightly greenish tint, and moreover becomes slightly granular, and,

[1] Ant. Schneider, who believes that these cavities communicate with the system of canals, describes the latter as bounded by a membrane, with nuclei and finely striated fibres, and calls these fibres " muscular (?)," loc. cit., pp. 8, 9.

[2] This it is which Ant. Schneider (loc. cit., p. 8) has described as stalked, and supposes them to be contractile !

therefore, to a certain extent opaque. This character belongs to it alone; under the influence of the same reagents, the amorphous matter in other parts of the body of Amphioxus remains hyaline, and the layers of fibres become stained bright red.[1]

In certain cases (fig. 5) a very clear and definite stratification of the amorphous matter may be seen in the papilla. The peripheral portion and the base are clearer and less opaque than the central portion, and the boundary between these different portions is clearly marked out. The central portion, which is also the darkest, rests on the layer of transverse fibres, and is raised up against the partitions of the cavities to where the endothelium is reflected.

In the centre of the papilla this dark zone presents a large projection, which itself may be surmounted by an intermediate zone.

V.—What histological and morphological signification can be attributed to the papilla and to the containing cavity?

The papilla evidently belongs to the category of connective or laminar organs.

The amorphous substances are everywhere very abundant in Amphioxus. In transverse sections (fig. 4) two thick layers (d, e) of greenish amorphous substance, hyaline and devoid of laminar fibres, may be distinguished on each side of the median aponeurosis. Elsewhere the amorphous substance is very dark (f, g), and contains thick fibres,[2] and although no part is so opaque as the papilla, it is evident that it is a simple variety of connective-tissue, characterised by the absence of cellular elements, the great scarcity of fibres, and lastly, by the probable presence of a certain quantity of fatty matter in a finely divided state.

The meaning of the cavity which surrounds the papilla is also evident. It is a space which is more developed than the rest, and which tends to take on the character of a serous sac. Silver nitrate gives the same result as on serous membranes; in the only preparation we have made, the network of silver staining is very clear upon the lateral walls of the cavities, but does not show upon the papilla, but no doubt could be obtained there also by employing suitable methods.

[1] After the more prolonged action of osmic acid and alcohol the papilla becomes darkly coloured, and certain coloured oily globules appeared to be emitted by it. This seems to indicate the presence of fatty bodies in its composition.

[2] Comp. 'Ant. Schneider, loc. cit., p. 7.

When one observes the gradations of structure between these cavities, the small separate cellular cavities placed at the extremity of the cord, the terminal cavities of certain rods, and, lastly, the rudimentary cavities in the swellings of the network in the neighbourhood of the mouth, one is led to regard these various structures as the first indications of a system of serous cavities and lymphatics. Besides this, the cellular lining of these cavities has quite the characters of the lining of the embryonic serous sacs in the higher Vertebrata. We confine ourselves to merely suggesting this as a probable analogy, as we have no exact account of the histogenic development of these parts.

Lastly, what morphological signification shall we give to the organ represented by the papilla and its cavity ?[1]

Here all terms of comparison leave us completely at fault; no homology whatever among vertebrates is to be thought of.[2] Their number corresponds neither with that of the spinal nerves, nor with that of the myotomes; they are about three or four times as numerous.

The connection which certain authors put forward[1] between the papilli and the fin-rays· ought rather to be considered as an *analogy* than a true *homology*. Amphioxus is not altogether devoid of either cartilage or bone. It is cartilage which forms the skeleton of the oral tentacles; it is bony substance which forms the framework of the branchial skeleton. The papillæ, looked at from the point of view of general anatomy, would form a third kind of skeleton element. They would represent a first stage in the differentiation of amorphous matter which is about to form a special organ.

It should not be forgotten that amongst Teleostean fishes a certain number of bony organs of the dermal skeleton result simply from a similar differentiation of amorphous intercellular substance, such as we have shown in the bony plates of Syngnathians. In the latter case, the amorphous matter becomes individualised as calcareous salts are deposited in it; in the papilli of Amphioxus fatty matters are no doubt deposited in it. In any case, both belong directly to a numerous and varied group of amorphous intercellular substances.

[1] We may here note a peculiarity which presents itself in the sheath of the notochord of Amphioxus. In the most successful transverse sections this presents above a slight median prominence (cf. Ant. Schneider, loc. cit., pl. xiv, fig. 1), and below two slight prominences on each side of the median line, which no doubt might be considered as the first rudiments of the vertebral skeleton.

[2] Anton Schneider, loc. cit., p. 8.

We point out this analogy here without giving to it more weight than seems justifiable, in the absence of special researches. We may add that, from whatever point of view we look at the subject, either that of comparative anatomy or that of general anatomy, a considerable difference still remains between the papilla of Amphioxus enveloped in its serous cavity and the dermal bony productions of fishes. Up to the present day, at any rate, intermediate conditions are wanting which would show how such a morphological evolution could have taken place as is taken for granted in bringing these organs together, from a more or less pronounced phylogenetic point of view, under a common denomination. All just ground for establishing a real homology between them is wanting. Although the papillæ partake, along with true fin-rays, of the character of skeletal organs, they clearly possess a special nature, without homologue, in other vertebrates.

Muséum de Paris, 1 June, 1880.

The PERIPHERAL NERVOUS SYSTEM *in* PALÆO- *and* SCHIZO-
NEMERTINI, *one of the* LAYERS *of the* BODY-WALL.   By
Dr. A. A. W. HUBRECHT, of Leyden.   With Plates
XXXII and XXXIII.

IN a paper, which was published in May last, in the
' Verhandelingen van de Koninklyke Akademie van
Wetenschappen te Amsterdam, vol. xx,' and which treats of
the anatomy and physiology of the nervous system of the
Nemerteans (an abstract of the paper with accompanying
plate appeared in the July number of this Journal), the
peripheral nervous system is dealt with very superficially
only. This was done on purpose. At the time when that
paper was prepared for the press the results which I had
arrived at concerning the peripheral nerves in the two sub-
orders of the PALÆO- and SCHIZONEMERTINI were so much
in contradiction with everything hitherto published on the
subject that I could not venture to publish these results
before I had been able to convince myself of their trust-
worthiness in numerous series of preparations, which should
gradually come under my observation. During the correc-
tion of the proof-sheets of that paper I was happily surprised
in obtaining, much sooner than I expected, different sets
of sections, which, by their superior state of preservation,
allowed me to settle the point. I could only mention this
briefly and insufficiently in the explanation of the plate be-
longing to that article. A more detailed account was at
the same time promised, and is now embodied in the follow-
ing pages.

A. *Central Nervous System and Cephalic Nerves.*

The central nervous system of the Nemertines is composed
of two longitudinal trunks running parallel to each other all
along the whole length of the body ; they are dilated ante-
riorly into a very simple or a more complicated pair of brain-
lobes, situated above the alimentary canal (often in front of
the mouth), and united together by a ventral and a dorsal
commissure, the latter always thinner than the former.
Posteriorly these longitudinal trunks either terminate very
close to the end of the body, or they are again united by a
third commissure, which, like the brain, passes *above* the
alimentary tract in the anal region. The whole of the cen-
tral nervous system (" marrow-trunks " and brain-lobes)

consists of fibrillar nerve-substance, to which a thick layer of nerve-cells is uninterruptedly applied. This cellular coating is nowhere dilated into ganglionic swellings, except in the brain itself.

An important difference is presented in the situation of the central nervous system with respect to the muscular body-wall in three different groups of genera. In the more primitively organised genera of the PALÆONEMERTINI (*Carinella annulata, inexpectata,* &c.) the *whole* of the nervous system lies directly under the epiderm *outside* of the muscles; in two other genera of PALÆONEMERTINI and in all the SCHIZO-NEMERTINI it lies enclosed *within* the muscular coat of the body, generally between the outer longitudinal and the inner circular layer of the muscles, whereas in the HOPLO-NEMERTINI the longitudinal marrow-trunks are situated *inside* the muscular body-wall in the body-cavity.

Three sets of cephalic nerves originate directly from the brain:—*a.* Nerves to the tip of the snout and to the eyes (if present). *b.* A pair of nerves for the proboscis. *c.* A pair of nerves for the wall of the œsophagus and alimentary canal (n. vagus).

B. *Peripheral Nerves.*

A rapid recapitulation of what is said about the peripheral nervous system of Nemertines by the three authors who have furnished the best original investigations on these worms—Quatrefages, Keferstein, and MacIntosh—may precede the enumeration of the results arrived at by myself, and, at the same time, serve to explain apparent discrepancies.

Quatrefages ('Annales des Sciences Naturelles,' vi, 1846, p. 278) says, speaking of the longitudinal marrow-trunks: "De ces troncs partent d'espace en espace des filets qui se portent probablement aux couches musculaires, mais que je n'ai jamais pu suivre assez loin pour être certain de ce fait. . . . . . Je n'ai distingué le long de ces troncs primitifs aucune trace de véritables ganglions. Seulement les filets qui en partent sont assez régulièrement épatés à leur base et quelquefois on pourrait croire qu'il y a là une sorte de renflement, mais cette particularité ne se réproduit pas d'une manière constante."

This is all the information he gives about the peripheral nervous system. I must insist upon the fact that all his figures (pl. 8, fig. 2; pl. 9, fig. 1) in which he represents the small branches springing from the longitudinal trunks belong to the suborder of the HOPLONEMERTINI. In a separate figure of the longitudinal nerve-trunks of *Carinella*

(pl. viii, fig. 3) *no* lateral branches are indicated in the drawing,

Keferstein writes ('Zeitschr. f. Wissensch. Zoologie,' Bd. xii, p. 80); "Aus den Seitennerven treten in regelmässigen Abständen (pl. v, fig. 10) feine Nerven, mit breiter Basis entspringend, aus, die ich nur bis auf unbedeutenden Abstand vom Seitennerven verfolgen konnte und die wahrscheinlich hauptsächlich in die Hautspgehen."

The figure in which he indicates these peripheral nerves implanted upon the longitudinal trunks is again, in this case, one of the Hoplonemerteans, viz. *Amphiporus splendidus*.

MacIntosh is the first to make separate mention of the state of things as he finds them in the armed and unarmed species.

Of the first he says ('A Monograph of British Annelids, *A. Nemerteans*,' p. 83) : "The branches given off by these trunks are generally pale and indistinct, but by the use of dilute acetic acid in *Amphiporus lactifloreus*, and in others without such aid, they can be satisfactorily observed.   They are easily seen, for instance, in *Amphiporus pulcher*, the reddish colour which tinges them at their commencement shining through the translucent integuments.   An elaborate plexus of branches from the lateral trunks has also been noticed in these species" (pl. xvi, fig. 3).

Of the unarmed species he writes as follows (ibid., p. 110) :

"Branches probably exist, but only faint traces of such are seen in the longitudinal sections, for the opacity of the textures in the living animal prevents their being satisfactorily made out."

MacIntosh's very hesitating affirmation concerning the presence of peripheral branches in the unarmed Nemerteans is certainly a step in advance of his predecessors, who did not notice any difference in this respect, who principally examined and only figured armed species, and regarded the facts as they found them in this subdivision at the same time as normal and typical for the whole order.   In this way the error has for a long time persisted, which I now hope to clear away definitely, that the peripheral nervous system of the Nemerteans corresponds in general to the type of the HOPLONEMERTINI, in which it was first described and figured.

My own investigations of the HOPLONEMERTINI did not furnish any new results worthy of note.   I found the small branches given off by the longitudinal trunks as the above-cited naturalists have described them.   These branches

follow each other at regular intervals, probably corresponding
in number with the successive internal metameric divisions
of the body. They remain undivided for a certain distance,
and, spreading out in the muscular tissue, are quickly sub-
jected to a dichotomic division, which gives rise to numerous
smaller twigs. They can be clearly made out by compres-
sion of the living animal, with or without the use of dilute
acetic acid, as well as in longitudinal and horizontal sec-
tions. I generally noticed two branches springing from the
same region of the longitudinal trunk, one turning to the
dorsal, the other to the ventral half of the muscular body-
wall (see Pl. XXXII, fig. 3, and in another paper, ' Aan-
teekeningen over de Anatomie enz. van eenige Nemertinen,'
Utrecht, 1874, pl. i, fig. 2). The elaborate plexus of
branches observed by *MacIntosh* is worthy of note, as I
will try to show hereafter ; it was sometimes observed by
myself, although in a lesser degree than is represented in
MacIntosh's figure.

The two other Nemertean suborders, the PALÆO- and the
SCHIZONEMERTINI, of which I examined several hundreds of
sections, most of them forming uninterrupted series, gradually
puzzled me more and more by the absence in all these sec-
tions, longitudinal as well as transverse, of any small branch
springing from the longitudinal marrow-trunks. Even the
faint traces which *MacIntosh* believes to have observed now
and then did not make their appearance, and the better the
sections were preserved and stained the more conspicuous
was their absence. On the other hand, well-preserved sec-
tions of Schizonemerteans always show the thick coating of
nerve-cells surrounding the central, more fibrous, tissue of
the longitudinal trunks very clearly (Pl. XXXII, fig. 6).
The whole of the trunk and cellular coating is again sur-
rounded by another layer of homogeneous connective tissue,
which separates at the same time the external layer of longi-
tudinal muscular fibres from the internal circular muscular
layer, not only in the vicinity of the nerve-trunks, but
throughout the whole extent of the body (cf. Pl. XXIII,
fig. 12, in the July number of this Journal). In this homo-
geneous layer is situated another somewhat thinner layer of
tissue of an entirely different histological character.

It is this last-named layer which we now have to ex-
amine in detail. Staining reagents, as picro-carmine, &c.,
have very little effect upon the bulk of the tissue, although
they render conspicuous in it the presence of numerous
nuclei, which distinctly stand out with a more or less deep red
tinge. Each nucleus is provided with a nucleolus, and now

and then traces of the circumference of a cell enclosing this nucleus are visible. No difference can be detected between these nuclei and those of the nerve-cells of the brain- and marrow-trunks. The bulk of the tissue which remains colourless has a finely punctate-fibrillar appearance, resembling in this respect the fibrillar nerve-substance as we find it in the centre of the longitudinal trunks of these worms. A characteristic difference is, however, noticed in the fact that the separate cells appear to be more numerous in this fibrillar plexus than they are in the fibrous centre of the longitudinal trunks.

This curious layer forms a cylindrical sheath throughout the whole body between the longitudinal and circular muscular layers. The median dorsal nerve (which in a former paper I proposed to call the nerve for the proboscidian sheath) appears as a simple thickening of this layer, and laterally the longitudinal marrow-trunks may in the same way be looked upon as parallel thickenings of the same tissue, as the ganglion-cells in the longitudinal nerve-trunks gradually merge, without any considerable difference in histological character, into the cells of this layer. In the sheath of ganglion-cells belonging to each longitudinal trunk the boundaries of the cells are, however, more conspicuous and always visible, and this is not so distinctly the case in the circular layers. This cylindrical tunic of distinct tissue, which in front is applied to, when not in direct communication with the tissue of the brain (Pl. XXXIII, figs. 9 and 10), must no doubt be regarded, after what we have noticed about its histological character, as *a nervous layer*. Its constituents are nerve-cells as well as fibrillar nerve-tissue.

Pl. XXXIII, fig. 12 shows a small part of this nervous tunic in a horizontal section through the back of the animal ; *nps.* is the median dorsal nerve ; *nl.* is the fibrillar plexus with the separate cells, forming together a layer, differing in thickness according to the size of the animal, and always only partly visible in a single section. The longitudinal (*LM.*) and circular (*TM.*) muscles are only sparingly indicated in the figure, as are also the thin fibres which directly traverse the nervous layer (cf. figs. 6 and 7).

The hypothesis of the nervous nature of this layer is strongly confirmed by processes which are sent out by it into the adjacent muscular tissue, and which generally carry one (fig. 8) or more nerve-cells (fig. 11) ; sometimes none. At other times these ramifications have a still more marked fibrillar appearance ; for example, those which leave the median dorsal

nerve and stretch vertically downwards towards the pro-
boscidian sheath. I believe I am justified in regarding these
processes as the equivalents of the smallest terminal twigs
of the nerve-branches in the HOPLONEMERTINI and other
animals with a dichotomically divided peripheral nervous
system. At the same time I must regard the nervous tunic
just sketched as representing *a more primitive type of peri-
pheral nerve-system, in which it has not yet come to a locali-
sation into transverse branches, metamerically placed, but in
which one of the layers of the body-wall is yet in function as
the recipient and conductor of nervous stimuli.*

The facts observed and above described for the numerous
species and genera of SCHIZONEMERTINI are so palpable and
unmistakable, and at the same time so constant,[1] that the
question immediately presents itself :—How do the less differ-
entiated and older types of Nemerteans, the PALÆONEMER-
TINI, and more especially the genus *Carinella,* behave in this
respect ?

Pl. XXXII, fig. 4 and 5, may serve to give the answer to this
question. We find the whole of the central nervous system
directly under the epiderm, exteriorly to the muscles of the
body-wall, as was noted above. A thick basement membrane
separates the cellular parts of the skin from the subjacent
muscular tissue, and in this basal membrane we find in
favorable preparations (those in which this basal membrane
is somewhat distended, being preferable) that the sheath of
nerve-cells (*nc.*) belonging to the longitudinal trunk is directly
continuous with a layer (*nl.*), which here again is provided
with nuclei of exactly the same character as those of the
nerve-cells and ensheathes the body *exteriorly to the muscles.*
The histological character of this layer corresponds to what
we have found in the nervous tunic of Schizonemerteans.
Here, again, a fibrillar structure and the scanty absorption of
staining reagents would suggest the idea of nervous-tissue,
even were the direct passage into the nerve-cells of the
lateral trunks less perfectly demonstrable. In front of
the brain this layer is continued in the head, being more
conspicuous ventrally and laterally than dorsally. Here,
moreover, the fibrillar structure is far more apparent, and
the enclosed nerve-cells are less numerous.

In *Polia* and *Valencinia* the same layer is present, only

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[1] The opinion on the nervous nature of this layer, first emitted in my
above-cited paper, 'Zur Anatomie und Physiologie des Nervensystems der
Nemertinen,' p. 47, has only lately been confirmed for another genus of
Schizonemerteans by Herr Dewoletzky of Vienna ('Zoologischer Anzei-
ger,' No. 62, p. 399).

enclosed between two muscular layers, as it was described for the Schizonemerteans, and, moreover, corresponding with the latter in the fact that this layer does not occur also in the head in front of the brain, as it partially does in *Carinella*. In all genera higher differentiated than the latter this plexus-like arrangement in the head (which I first mistook for a considerable number of parallel nerves, on account of the appearance in transverse sections) appears to have developed into a smaller number of distinct nerve-stems, dichotomously dividing and innervating the tip of the snout, the muscles of the head, and the eyes, when present.

The fact of the presence of the nerve-sheath in *Carinella* on the outside of the muscular body-wall and under the immediate covering of the ectoderm is of the more importance, as we have indeed to look upon this genus as one of the less differentiated of all Nemerteans[1] which must serve as a starting-point for comparisons by which to determine the morphological significance of any structure which is common to this and other genera.

In the preparation figured (Pl. XXXII, fig. 4) I find that processes (*np.*) are being sent out from this layer as they were described for the Schizonemertini. Here I only succeeded, however, in demonstrating such as were turned towards the muscular layers; in other sections I find that processes towards the epidermic cells are also present (fig. 5). As the nervous layer is generally closely applied to the muscles, it is difficult to observe the centripetal processes, and only in such a case as the one figured, where by some reason or other this layer lies somewhat removed from the subjacent muscles, can these important ramifications be noticed.

The diagrammatic figures 1, 2 and 3, will now be more intelligible after this rapid exposition of the facts. Fig. 1 applies to *Carinella*. The lateral trunks and the nervous layer are wholly *outside* the muscular body-wall, processes from this layer being sent outwards and inwards. Fig. 2 stands for *Valencinia, Polia,* and all the SCHIZONEMERTINI. The trunks and the layer are enclosed *within* the muscles, the peripheral processes are sent out in both directions. Fig. 3 finally represents the HOPLONEMERTINI, a stage of higher development, in which the continuous cellular layer uniting the longitudinal trunks and ensheathing the whole body has become localised and differentiated into separate nerve-branches springing from these trunks at regular intervals.

[1] Cf. A. A. W. Hubrecht, "The Genera of European Nemerteans critically revised," 'Notes from the Leyden Museum,' vol. i, p. 19.

That a tendency to a metameric arrangement of the other-
wise indifferent layer is nevertheless already present in the
SCHIZONEMERTINI is shown in fig. 13, which illustrates a
thin section through the longitudinal muscular layers, im-
mediately above the nervous sheath. Three of the peripheral
processes taking their course through interstices between
the muscles have been cut in this section; they are
regularly arranged along a transverse row, and numerous
other sections show that indeed an approach to regularity
may in this respect be noticed all over the body. If this
regular arrangement gradually combines with a thickening
of the nervous layer along the points from whence these
processes originate, and with a corresponding reduction along
the intermediate spaces, the formation of transverse indi-
vidual nerves (which at first would have the appearance
of commissures,[1] and in a later stage would take the aspect
of transverse branches springing from the longitudinal
trunks), carrying perpendicular smaller twigs, would be the
inevitable result. And it is in this way that we must
form our conceptions of the intermediate stages lying
between *Carinella* and the HOPLONEMERTINI. As such must
also be regarded the plexus-like arrangement between two
successive peripheral branches, as observed by *MacIntosh*
and noted above.

A certain variability in the aspect of the nervous tunic in
small and large specimens, which, though generally a con-
tinuous layer (even if not of considerable thickness), on
other occasions has the appearance of a plexus with very
narrow meshes, confirms the supposition that local varia-
tions of the thickness of this layer, finally leading to the
formation of separate nerve-stems, may occur. Generally
bundles of radial fibres (*rf.*), which in other places are more
isolated, take their course through these meshes.[2]

[1] In a recent paper of von Kennel's ('Die in Deutschland gefundenen
Land-planarien,' Würzburg, 1879, p. 39) he speaks (quite aphoristically)
of transverse commissures uniting the longitudinal trunks of Nemerteans
across the back of the animal. It may be that what he took for commis-
sures are portions of the nervous sheath above described; still it appears
improbable that this excellent observer would have overlooked the nervous
tissue which unites the longitudinal trunks ventrally as well. Surface
sections (Pl. XXXIII, fig. 12) show that this questionable arrangement of
the tissue in transverse commissures, now and then met with in longitudinal
sections, is merely apparent.

[2] When I showed some of my preparations to Prof. E. Ray Lankester
last April, he drew my attention to the apparent agreement between this
nervous layer and Stricker's nerve-layer of the epiblast of the embryo-
frog. I am not disposed to believe that any deep significance can be
attached to this rather distant resemblance; but the parallelism, such
as it is, is interesting.

Another question which future investigations will have to solve, and which at present I can no more than briefly call attention to, is this—How are we to conceive the nervous sheath to have gradually become enclosed between the muscular layers? More especially can the difference between *Carinella* and the Schizonemerteans be explained? If I venture to emit an hypothesis at present for which I cannot as yet furnish sufficient or conclusive evidence, and which presented itself to me in studying the beautiful investigations of *O.* and *R. Hertwig,* on the structure of the *Actiniæ* ('Jenaische Zeitschrift,' 1880), it is because it appears at the same time to explain the fundamental difference in the muscular system of different groups of Nemerteans, which was first clearly pointed out by *MacIntosh.* The hypothesis might be briefly formulated thus : the external longitudinal muscular layer in *Polia, Valencinia,* and all the SCHIZONEMERTINI must be regarded as a special development of the ectodermal musculature." If this be true (and the curious arrangement of the external longitudinal muscles in *Polia* compared with *Carinella,* on the one hand, with the SCHIZONEMERTINI, on the other, appears to furnish strong arguments in its favour), the wandering inward of the nervous system can be more clearly understood, those muscles, which from the first were situated exteriorly to it having gradually attained this strong development and fascicular arrangement. The inner longitudinal and the circular muscular layer, as they are present in *Carinella* and the HOPLONEMERTINI, would thus represent the typical muscular system of Nemertine organisation, whereas the exterior layer of longitudinal muscles, which is added to it in the SCHIZONEMERTINI, *Valencinia,* and *Polia,* would appear to be of accessory character.

The curious distribution of nervous tissue above described is, I believe, of importance towards the solution of two problems touching the physiology of these worms.

These problems are—1st. The extraordinary restorative power of a great number of Nemerteans. 2nd. Their intense sensitiveness to outward stimuli, which may even occasion spontaneous rupture of the animal into two or more pieces.

As to the restorative power, it has often been noticed (*Dalyell, MacIntosh, Barrois*) : and detailed observations are on record of severed portions of the trunk having regained a complete central nervous system—brain, respiratory lobes, and eyes included—when only a cylindrical fragment, open at both ends, was left. The tail end is always sooner restored, but under favorable circumstances the restoration of the head

and brains does not seem to be rare. Although I never had occasion to study the phenomenon myself, the descriptions of *Barrois* and *MacIntosh* do not leave any doubt as to the way in which it progresses. Three months after the fragment was separated from the parent it was again provided with the normal brain-lobes and all their accessories, which had developed at the anterior ends of the nerve-trunks after the cicatrising process at both ends of the fragment had been concluded. Another fragment was severed from the parent animal in September, developed perfect spermatozoa and normally deposited them in February next.

Looking upon the lateral trunks as local thickenings of the more indifferent and primitive nerve-sheath in the body-wall, these curious facts appear in another light. For if we have succeeded in demonstrating that nervous tissue, which persists in a more primitive stage, the cellular elements being scattered indiscriminately among the fibrillar, is diffused throughout the whole circumference of the body-wall, then the appearance of local thickenings in this tissue finally leading to the formation of lateral trunks, and eventually of brain-lobes *sensu strictiori*, is less startling than would be the reproduction of the central apparatus out of lateral trunks, which hitherto have been regarded as essentially peripheral.

I must here lay stress upon the fact that in the HOPLO-NEMERTINI, where this primitive nerve-sheath has already been replaced by separate diverging peripheral branches, and, which, therefore, have attained a far more specialised stage respecting the structure of their nerve-system, no case of similar reproduction of the head and brains has ever yet been noticed, as far as I know.

All such observations were made upon SCHIZONEMERTINI, and I do not believe it will prove a hazardous prophecy to predict that later investigations will show that in the armed species it really never occurs. At the same time the latter die much quicker after having been severely injured than do the Schizonemerteans, the families of *Amphiporidæ* and *Tetrastemmidæ* enjoying a still more restricted longevity than do the family of *Nemertidæ s. str.*

All this appears to me to be to a great extent explained by the above described fundamental differences in the distribution of the nervous tissue in the body-wall.

As to the extreme sensitiveness to outward stimuli, it has been noticed by all former observers, and *Leuckart* applied the name of *somatotomus* to one of the species, because it regularly broke itself into pieces upon being handled or touched.

This sensitiveness, with which the SCHIZONEMERTINI are none the less endowed than the HOPLONEMERTINI, would appear the more remarkable in the former in the absence of peripheral branches springing from the longitudinal marrow-trunks. The presence, however, of a sheath of nervous tissue in every region of the body, either immediately under the epiderm or inside the muscles, and sending out processes as well to the exterior surface as to the internal muscular layers, is quite as sufficient an explanation for this extreme sensitiveness as we before showed it to be for the persistence of life in the several fragments, which even developed normal spermatozoa months after they had become separated.

As will be understood from the contents of the foregoing pages, my investigations into this curious distribution of nervous tissue in the body-wall of the Nemertines are far from being either complete or exhaustive. Not having as yet had occasion to examine fresh specimens with special regard to this nervous layer, I must reserve the more delicate histological questions for a future publication. The direct connection between the fibrillar plexus and the exterior cellular layer of the body, the presence of any distinct sensory cells in this ectodermic layer, as well as the connection with the muscular fibres, on the other hand—all this can only be studied in careful preparations, teased out from the tissues when quite fresh, according to the delicate methods which have, for example, been so successfully applied by *O.* and *R. Hertwig,* in their splendid researches upon the nervous system of the Cœlenterata ('Das Nervensystem und die Sinnesorgane der Medusen,' and further 'Die Actinien, anatomisch-histologisch mit besonderer Berücksichtigung des Neuromuskelsystems,' 1878-80).

Having as yet only had series of sections mounted in balsam at my disposal, I could not expect to penetrate into all these histological details merely by their aid; and if, nevertheless, I venture to publish these preliminary results it is because they appear to me to possess an increased interest now that the researches above mentioned of *O.* and *R. Hertwig* have come before the public. The very lowest stage of differentiation in which nervous tissue is at present known in the animal kingdom was found by these searchers[1] in the Actinia, where it appears as a fibrillar plexus, with nerve-cells indiscriminately scattered in it

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[1] Might not this word take the place of the barbarous "*savants*" so often met with in English scientific papers of the present day? It would correspond to the German "Forscher."

throughout a great part of the ecto- and entoderm.   In the oral plate (Mundscheibe), where the nerve-cells are more numerous and situated closer to each other, there is a tendency towards a certain degree of centralisation.

It cannot be denied that this primitive arrangement offers strong analogies to the nervous plexus above described for the Nemertines, which, on the other hand, approach, in the histology of their nervous system, in many respects to what *A. Lang* has so carefully described for other PLATYELMINTHES (' Untersuchungen zur vergleichenden Anatomie und Histologie des Nervensystems der Plathelminthen.' 1. Das Nervensystems der marinen Dendrocoelen.   2. Das Nervensystem der Trematoden; Mitth. aus der Zool. Station zu Neapel, 1879-80).   And so I thought that giving my results, even in their incomplete form, might be suggestive of further researches into these questions; the more so because, in a former paper (' Verhandelingen v. d. Kon. Akad. v. Wetensch. te Amsterdam,' vol. xx, 1880; cf. July number of this Journal), I insisted upon the phylogenetic importance which the nervous system of the Nemerteans appears to possess when compared to that of the ANNULATA, on the one hand, and of the VERTEBRATA, on the other.

In the monograph on this group of worms, which I have now in preparation for Professor Dohrn's series of publications, illustrating the Fauna and Flora of the Bay of Naples, I hope to be able to furnish evidence about those points in which the present paper confesses to be deficient.

The EYE *of* PECTEN. By SYDNEY J. HICKSON, B.Sc.,
Scholar of Downing College, Cambridge. (With Plates
XXXIV and XXXV.)

THE general absence of organs of vision amongst the
members of the class Lamellibranchiata meets with a curious
and interesting exception in the genera Pecten and Spon-
dylus.

These genera have long been known to possess a great
number of eyes of considerable complexity, situated on the
border of the mantle. The number of these eyes varies
considerably in different individuals, ranging in the genus
Pecten from eighty to one hundred and twenty. Their
position also varies ; for, although they are always situated
on the border of the mantle, yet sometimes they are placed
at equal distances from one another, and sometimes they are
clustered together in certain localities.

Notwithstanding this indefinite element, both in their
number and position, which might be expected to run
parallel with a primitive and simple organisation, their
anatomy is exceedingly complicated, and exhibits all the
most important structural elements of the eyes of the higher
Vertebrata.

The earliest investigations into the anatomy of Pecten's
eye are those of Krohn,[1] who gives a drawing of the course
of the optic nerve. This drawing is copied in many of the
subsequent papers on the subject by other investigators, and,
as far as it goes, is correct. Duvernoy,[2] in his description
of the nervous system of the Pectens, gives a short descrip-
tion of the anatomy of the eye. This paper, however, is
chiefly valuable for the excellent figures and descriptions
of the distribution of the nerves in the mantle, and the
filaments which are given off from the main trunks of these
to supply the tentacles and the eyes.

The researches of Blanchard[3] and of Keferstein[4] which
followed did not add very much to our knowledge on this
subject, and it was not until 1865 that any careful histo-
logical inquiries were carried on. It was Hensen[5] who first

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[1] Krohn, 'Müller's Archiv,' 1840, p. 301, pl. xi.
[2] Duvernoy, 'Mémoires de l'Academie de Sciences,' t. xxiv, 1852.
'Mémoire sur le système nerveux des Acephalés,' p. 73, pl. ii.
[3] Blanchard, 'Organisation du regne animal : Mollusques Acephalés.'
[4] Kerferstein, 'Zeit. für wiss. Zoologie,' 1863, p. 133.
[5] Hensen, 'Zeit. für wiss. Zoologie,' 1865, p. 220.

gave figures of the characters of any of the histological elements. But as the eye of Pecten forms only a very small part of the paper, his figures and description are by no means complete, and in many respects they are incorrect. Finally, J. Chatin[1] has contributed two short papers, without figures, on this subject.

Of the scanty literature Hensen's paper is by far the most important, and he alone gives any good figures of sections of the eye, or of its elements ; the other observers give remarkably few figures, and consequently I have had, owing to an imperfect knowledge of the German language, some difficulty in making myself acquainted with the substance of their papers.

I have been encouraged in publishing the following researches chiefly by this scarcity of good figures, but also because I believe, and will give my reasons for believing, that these eyes deserve more mention than is usually made of them in our zoological text-books.

My investigations were chiefly carried on upon *Pecten maximus*, but I have also had the opportunity of making sections of and studying the eyes of two other species, *Pecten jacobæus* and *Pecten opercularis*. The eyes of these three species differ from one another in one or two not altogether unimportant particulars, and, as I shall afterwards point out, they form an interesting gradation, the points of difference between *P. maximus* and *P. opercularis* passing through intermediate stages in *P. jacobæus*.

The eyes of Pecten maximus—are situated amidst a number of tentacles, which run all round the border of the mantle. These tentacles are capable of considerable movement, and frequently overhang the eyes and protect them from the light. The eyes themselves are situated upon short stalks, which resemble very closely the basal part of an ordinary tentacle.

This similarity caused Duvernoy to name a tentacle a tactile pedicel, and an eye an ocular pedicel, thus to a certain extent implying that they are morphologically homologous organs respectively modified for a tactile and an ocular function. This homology is justified by certain points in their anatomy, such as the course of the nerve and the arrangement of the muscular fibres, and I believe that when the development of these eyes is studied the homology will be still further confirmed.

The border of the mantle which bears the tentacles and

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[1] J. Chatin, 'Bulletin de la Société Philomatique.' Paris, 1877.

eyes is covered with an epithelium, consisting of columnar, non-ciliated, and slightly granular cells bearing nuclei, situated near the base of the cells. As this epithelium passes over the eye-bulbs, it undergoes two interesting modifications. It becomes considerably thicker and filled with a dark brown pigment (Pl. XXXIV, fig. 1 c) as it passes round the sides of the eyes, but immediately in front of the eye (Pl. XXXIV, fig. 1 a), it again diminishes in thickness, and becomes perfectly transparent. By thus surrounding the eye on all sides with a dark-coloured pigment, leaving only a round spot in front, clear and transparent, the epithelium, by limiting the entrance of the light to a small diaphragm in front, here performs the function of an iris. The epithelium which runs over this transparent part, and which forms the epithelial layer of the cornea, differs from the ordinary epithelium covering the rest of the mantle in that their cells are rather larger, are perfectly transparent in the living condition, and the nuclei are large and spherical, and situated in the centre of the cells.

The eye consists of the following parts, which I shall now describe in order. The cornea, covered externally by its transparent epithelium, protects a large elliptical lens. Close up to the lens is the retina, but separated from it by the optic nerve, which spreads out over the anterior surface of the retina. The retina rests upon a tapetum, and behind this, occupying all the posterior concavity of the eye-cup, there is a red pigment.

The *cornea*—consists of two parts, the outer epithelium, which has already been described, and a basement membrane, consisting of a thin layer of connective tissue. As before stated, this epithelium is merely a modification of the general epithelium of this part of the mantle ; and the pigmented epithelium surrounding the eye-bulbs (in like manner, a modification of the same tissue) is continuous with it all round its edge. The passage of the cells of the pigmented epithelium into those of the corneal epithelium is signalised by two important changes in the characters of the cells. In the first place the pigment entirely disappears, and the nuclei, which in the former case were obscured by the pigment, now becomes apparent, and in the second place the cells are considerably diminished in their longitudinal axis. The diminution in size of the cells causes the edge of the cornea to be sunk below the level of the pigmented epithelium ; and a shallow trough runs round the line of its juncture with it (Pl. XXXIV, fig. 3). The convexity of the cornea is not great, and the dome of it frequently only just

reaches the level of a line drawn from the highest points of the pigmented epithelium on either side of it. This appearance is not often seen in sections, as the pigmented epithelium rapidly shrinks, when the tissue dies, and under most reagents ; but I am fully persuaded of the accuracy of this statement from an examination of the eyes of living specimens of *Pecten maximus* and sections of *Pecten opercularis*.

The delicate epithelial cells of the cornea, in consequence of being entirely unprotected by any membrane similar to the conjunctiva of the higher animals, are quite naked, and very liable to injury from the rough edges of the tentacles which surround them. The arrangement just described, however, probably prevents the tentacles from coming into immediate contact with them. The little trough which runs round the margin of the cornea always contains a little liquid, even when the eye itself is removed from the water ; and the pressure of the tentacles when folding over the eye causes it to spread out as a thin layer over the cornea, and thus the cells are prevented from coming into immediate contact with the tentacle.

Thus, the two remarkable modifications, namely, the presence of a large quantity of pigment, and a greater longitudinal axis of the cells which the pigmented epithelium exhibits, are of considerable value to the eye, firstly, to prevent very divergent rays from entering, and secondly, to prevent any damage to the cornea caused by the rubbing of the adjacent tentacles over the sides of the eye.

The second layer of the cornea is about half as thick as the epithelial layer, and, like it, is perfectly colourless and transparent. It consists merely of a thin continuation of the connective tissue of the stalk. It may be called the basement membrane of the corneal epithelium, as from the absence of any definite cellular elements its only function probably is to support these cells.

Beyond the cornea this membrane becomes much thicker, and supports the pigmented epithelium, and at the same time structural elements make their appearance in it. From thence it passes into the connective tissue of the eye-stalk without further modifications.

The *lens*—is one of the most interesting parts of the eye. It is comparatively large, and is composed of a number of nucleated cells. In the fact that the lens is formed by more than one cell the eye of Pecten bears an interesting resemblance to that of the Vertebrata. The shape of the lens has been a subject of much dispute amongst the authors

who have written on this subject. Krohn and Keferstein believed it to be spherical. Hensen has figured it filling up the space between the cornea and retina, and consequently of an irregular bi-convex shape.

It is difficult to see how a controversy on such a simple subject could have arisen, unless it is because different authors have examined different species, and described them for the genus.

As regards *Pecten maximus,* an examination of the fresh eye has convinced me that in this species the lens is elliptical, the major axis being parallel to the plane of the mantle. A section of the eye made in a plane at right angles to the plane of the mantle and the direction of its margin—that is, the plane which is most convenient for section-cutting, and the one which is apparently usually adopted—would consequently cause the lens to appear circular in section. In the diagrammatic representation of the eye (fig. 1) I have for convenience sake represented the lens as being at right angles to the plane of the mantle in order that the true shape of the lens may not be overlooked.

A fresh examination of the lens, when teased out from the rest of the eye, exhibits one or two interesting points. The lens is not, as in most eyes, perfectly colourless, but possesses a well-marked brown colouration, and a number of fine striæ may be seen running in the direction of the major axis. The lens does not appear so perfectly elliptical in the fresh condition as in certain sections I have made; it is drawn out somewhat longitudinally, so as to be more like a double cone than an ellipse. This is probably due to the lens being released from the ligaments and connective-tissue pressures, which cause it to retain its proper shape.

Hensen says that the lens is very soft, and the cells are light, polygonal, and nucleated. A careful examination of the lens of *P. maximus* has led me to a very different conclusion. The lens seemed to be of exactly the same nature as in the higher forms, and when teasing it out I found some difficulty in holding it with a needle, as it slipped away from under it when a slight pressure was exerted. As regards the shape of the cells composing the lens, they are not all polygonal, as would be inferred from Hensen's remarks on the subject. In the centre they are polygonal, but as they approach the periphery they become more and more flattened and elongated, until at the periphery they are strap-shaped. They are nucleated. As Hensen, I could find no membrane covering the lens, and no muscular fibres connected with it; but in a few cases I have observed a liga-

ment, such as I have represented diagrammatically in fig. 1 *f*, which, I believe, forms a support for the lens. This ligament is usually broken by the action of reagents, and then hangs down by the side of the cavity, and thus becomes difficult to observe; at the same time the lens sinks down, and rests upon the anterior surface of the retina.

The lens is suspended in the space which corresponds with the vitreous humour in the higher animals. This space is filled with an aqueous humour in Pecten. The lens is larger, and, consequently, the space occupied by aqueous humour relatively smaller in *P. maximus* than it is in either *P. jacobæus* or *P. opercularis*, and in *P. jacobæus* it is larger than in *P. opercularis*.

The *retina*—does not line the concavity of the eye-cup, as it does in most well-developed eyes, but is nearly flat, and a considerable space is left between it and the floor of the cup, which is filled up by the red pigment. In consequence of this the retina appears in section to be a thick band crossing the eye from side to side. Thus, just as the lens was remarkable for the way in which it approached the retina by hanging back into the cavity, so the retina is remarkable for the manner in which it leaves the posterior concavity of the eye-cup to approach the centre. The eye of Pecten, in fact, presents the interesting peculiarity of the approach of the lens and the retina towards the centre, so that in *P. maximus* they almost touch.

The anterior surface of the retina is convex at the sides and concave in the middle, but these convexities and concavities vary in different species. The different layers of the retina will be described from behind forwards, as it will be easier to trace the transitions in that way than if described from before backwards. They are—1°. Posterior limbs of the rods. 2°. Anterior limbs of the rods. 3°. Spindle-shaped nucleated rods. 4°. Molecular and nuclear layer. 5°. Nerves.

The posterior limbs of the rods stand upon a membrane, which runs along the posterior side of the retina; at their anterior ends they pierce a very delicate membrane, and pass into the anterior limbs of the rods. The anterior limbs are about twice as long as the posterior limbs, and are usually smaller in diameter, and situated farther apart than the posterior limbs. That they are circular in section may be seen from Pl. XXXV, fig. 7*a*, which is a drawing of a section made at right angles to the eye-stalk. The anterior limbs of the rods are sometimes swollen so as to appear oval;

this condition occurs especially in the rods at the side con-
vexities. Fig. 6 represents an isolated rod in this con-
dition.

The anterior ends of the rods contract considerably, and
again expand into spindle-shaped bodies, each of which con-
tains a nucleus; so that in *P. jacobæus*, where the retinal
elements of this region are difficult to distinguish, there
may be seen a single row of nuclei running from end to end
of the retina, and following its sinuosities (Plate XXXV,
fig. 11 *b*).

In some of the rods at the side of the retina a second
spindle-shaped body follows the first one, as represented in
the isolated rods in figs. 5, 6, but usually the anterior end
of the spindle is drawn out into a delicate thread, which
occasionally possesses nuclear swellings. Finally, this thread
breaks up into a network, which bears a number of nuclear-
like bodies at its nodes, and several round molecular bodies
appear to be caught in its meshes. These bodies are so
much like the ordinary nuclei of the network that I am in-
clined to believe that they are, in reality, merely modifica-
tions of them, and in some way connected with the network
(fig. 6 *a*). Anteriorly the fibres of the network bend at right
angles and enter the nerve layer, which covers the anterior
surface of the retina. This nervous layer will be described
with the description of the optic nerves.

The above is a description of the retina as I found it in
P. maximus, and I believe it holds good for the other mem-
bers of the genus. The elements of the retina are so much
larger in this species, and the spaces between the rods and
network, &c., so much more considerable, that it is a great
deal easier to investigate; but I believe careful examination
of the other species would show that they do not differ from
this in any important detail.

The *tapetum*—is placed immediately behind the retina, and
may help in its support. When fresh,[1] the tapetum exhibits
a display of colours, and it is this membrane which gives
the eyes their beautiful metallic lustre. When examined
with a $\frac{1}{5}$th-inch obj. it seems to be composed of a great
number of little black specks separated by a fine yellow
membrane, but careful examination with a higher power
shows that it is composed of a great number of fine fibrils
crossing at right angles.

The space between the tapetum and the posterior part of

[1] I have one series of sections stained in osmic acid, and mounted in
Canada balsam, which has retained this display of colours.

the eye-cavity is filled with a red fluid *pigment*. In the fresh condition the pigment readily flows on to the slide when the eye is pricked, but in sections of the eye which has been hardened by alcohol or other reagents the pigment adheres to the tapetum or posterior wall of the eye-cup.

Hensen figures a layer of cells in this position, but I have never been able to observe anything of the kind; the pigment contains no cellular elements at all, nor is there a layer of cells lining the cavity which contains the pigment. The pigment consists of a number of bright red granules floating freely in a colourless fluid.

The *nervous supply*—of the eye of Pecten is perhaps the most interesting of the many peculiarities of this eye. The nervous system of Pecten is well described by Duvernoy in the paper referred to above. The mantle is supplied by a number of branches given off from the principal ganglia. These branches all fall into a large nerve, which runs round the margin of the mantle, and which Duvernoy calls the " circumpalial" nerve. This nerve is figured in section in fig. 1, Pl. XXXIV, one of the nerves joining this nerve being figured at fig. 1, *q*. This "circumpalial" nerve gives off filaments to supply the tentacles and eyes.

Krohn first gave a drawing of the optic nerve, and described it as a single nerve passing off from this trunk, and dividing into two branches as it aproaches the eye. Later observers have, however, drawn and described two nerves passing off from the "circumpalial" nerve. My researches have led me to believe that Krohn is right, and that such a figure as Hensen gives in his paper, representing two main trunks passing up to supply the eye is erroneous. Plate XXXV, fig. 9, of *P. maximus*, shows the division of the single nerve into its two branches. In fig. 1 the course of the optic nerve, before its division into two branches, was carefully drawn from one of a complete series of sections, and in none of the other sections could I find a trace of any other nerve proceeding from the "circumpalial." The branching of the nerve takes place in a plane at right angles to the plane of the mantle. When the optic nerve approaches the eye it divides into two branches, which may be called the " retinal nerve" and the " complementary nerve." The former passes up the side of the eye cavity, and spreads over the anterior surface of the retina ; the latter soon loses its sheath, and divides up into a number of branches, which supply the tissues surrounding the eye. The course which the retinal branch takes may be seen in Pl. XXXIV, fig. 1, and in Pl. XXXV,

figs. 8 and 9. In figs. 8 and 9, the first section is cut through the optic nerve, and shows the manner in which the retinal branch runs up the side of the eye-cavity; the second section shows the manner in which the branch bends over on to the retina and spreads out. The distribution of the complementary branch is diagrammatically represented in fig. 1 *n*; it seems to divide into a number of branches which envelope the eye-cup, and probably send filaments to the cornea, lens, tapetum, and epithelium.

Comparison of the eyes of the three species, *P. maximus*, *P. jacobæus*, and *P. opercularis.*—The eye of *P. maximus* is undoubtedly the most highly developed, the eye of *P. opercularis* is the simplest, whilst *P. jacobæus*, although more like *P. opercularis* than *P. maximus*, shows many points in which it is intermediate between the two.

The lens in *P. opercularis* is separated from the retina by a considerable space (Pl. XXXV, fig. 10), and consequently the chamber containing the humour is relatively large. In *P. jacobæus* the lens is larger than in *P. opercularis*, and the chamber consequently smaller; and in *P. maximus* the lens is very large, and nearly touches the retina, the chamber of the eye being sometimes very small. A gradation is thus observed in the character of this part of the eye in the three species. In *P. maximus* but a small space is filled with humour, in *P. jacobæus* a much larger space is filled with it, and in *P. opercularis* there is a larger space still.

Again, when the retinas of the three species are compared, a similar gradation is found. The retina of *P. opercularis* is comparatively thin, and the concavity and convexities of its anterior surface slight. In *P. jacobæus* the retina is decidedly thicker, and the anterior surface is more convex at its sides than in *P. opercularis*; moreover, it may be noticed that the delicate membrane which separates the anterior from the posterior limbs of the rods has become bent up in the regions corresponding with the anterior convexities of the retina. In *P. maximus* all these variations become much exaggerated. The retina is much thicker than in either of the other species; and the side convexities of its anterior surface are much bolder (Pl. XXXV, fig. 11, *a, b, c*). The anterior concavity does not undergo much variation.

The shape of the membrane separating the anterior and posterior limbs of the rods is greatly altered. In *P. opercularis* this membrane is observed, in section, to stretch from side to side without any well-marked curves; in *P. jacobæus* two well-marked curves, corresponding with the anterior con-

vexities of the retina, are observed; but in *P. maximus* these curves are converted into two distinct folds, which run up into the substance of the retina. The membrane between the folds does not sink again as low as it is at the commencement of the folds, and consequently the central part of the retina is raised in the form of a table above the level of its sides. This elevation of the central part of the retina may be also seen in *P. jacobæus*, though it is not nearly so well marked. The folds which occur in *P. maximus* cause the rods to appear to be given off in a pinniform manner at the sides of the retina, and before I found the intermediate condition in *P. jacobæus* I had some difficulty in determining the true relationship between the retinas of *P. maximus* and *P. opercularis*. (Compare *a, b, c,* fig. 11).

In addition to those just mentioned there are other minor points in which the eyes of these species differ from one another, such as in the shape of the cells composing the lens and in the distribution of the retinal nerve, &c., but they are comparatively slight.

General considerations.—Having thus described, in some detail the anatomy of the various parts which compose the eyes of Pecten, I shall, before leaving the subject, point out some of their interesting morphological peculiarities. It is, in itself, a remarkable thing to find a large and variable number of eyes situated on an area at some considerable distance from any central nerve-ganglion; and, when it is remembered that the class and even family (with one other exception, *e. g.* Spondylus) to which the genus belongs, possess no organs of vision at all in the adult condition, it is altogether surprising that they should be of such extraordinary complexity as they have proved to be. The high structural development that this eye has attained is, however, not so remarkable as the fact that in many ways it differs from the ordinary Invertebrate eye, and resembles that of the Vertebrata.

In the first place, the lens is built up of a large number of distinct nucleated cells, which undergo a flattening at its circumference very similar to that found in the eye of the Vertebrata. Whether the lens is developed from the cells of the epiblast, as in the Vertebrata, or from the mesoblast, must at present be left unsettled, but it will probably be found, when the development of the eye is studied, that in this respect also it resembles the eyes of the Vertebrata. The tapetum, a structure which is of considerable im-

portance to animals which are nocturnal or aquatic in habit, has hitherto been described only in the Vertebrata. That Pecten possesses a tapetum as highly developed as any found amongst the Vertebrata is anatomically a point of considerable interest; but it also indicates to a certain extent the physiological capability of the eye.

The chief interest, however, lies in the relative positions of the optic nerve, the retina, and the pigment. In the eyes of the Cephalopods the pigment layer is situated in front of the rods, and the nerve-fibres enter the rods from behind. In the eyes of the Gasteropoda, the Crustacea, &c., down to the simplest form of eye, such as that of the Rotifera, the same relationship of these parts holds good. In the Vertebrata, however, their relative positions are reversed; the optic nerve pierces the retina, and distributes itself over the front of the retina, whilst the pigment is situated behind it. In Pecten the relationship of these parts is the same as that in the Vertebrata; the nerve passing up the side of the eye-cup bends over, and spreads itself over the anterior surface of the retina. The pigment also is situated behind the retina. Pecten is not, however, the only Invertebrate whose eyes are built up on this type. Semper[1] has recently pointed out that on the backs of certain slugs (Onchidium) a number of eyes are found, and that in these the nerves pass to the front of the retina before being distributed. On account of this distribution of the optic nerve he says that they belong to the Vertebrate type of eye (typus der Wirbelthieraugen), so that two animals are now known, each belonging to a large and important class of Invertebrata (Gasteropoda and Lamellibranchiata respectively) that possess eyes which are built up on this type. The eyes of Pecten are even more deserving of the name of Wirbelthieraugen than those of Onchidium, for they are much more highly developed, and possess, in addition to this relationship of the nerve and retina, other Vetebrate peculiarities. The lens is multicellular, a character which, although not unknown amongst the Invertebrates, is much more characteristic of the Vertebrata. The tapetum, too, a structure which doubtfully exists in any other Invertebrata is found in Pecten and some Vertebrates. But, although the application of this word Wirbelthieraugen to these eyes is convenient for the adult condition, it must be carefully remembered that the development of these eyes is essentially

[1] Semper, 'Uber sehorgane vom Typus der Wirbelthieraugen auf dem Rücken der Schnecken.' Wiesbaden, 1877.

different from that of the Vertebrate eye. The Vertebrate eye is formed in the embryo from a hollow process given off from the brain, and the future eye-cup is formed by an invagination of this process. It is impossible for the eyes of Pecten or Onchidium to be formed by any process similar to this. Thus, in the young state these eyes are essentially different from those of the Vertebrata, and the resemblance in the adult is merely accidental, and by no means due to morphological identity.

Little is known and little can be said concerning the function of the eyes of Pecten. The presence of such a well-formed tapetum makes it probable that they are capable of appreciating very diffused light, and the close approximation of the lens to retina makes it exceedingly improbable that any image is formed upon the latter.

A few experiments have been made on the extent of their visual power, which make it very doubtful whether they are of much value to the animal in avoiding its enemies. The most reasonable theory of their function seems to be that, when on the ebbing of the tide, a probability arises that they will be left high and dry on the shore, they can appreciate the fact by the growing intensity of the light, and, by that peculiar flapping motion of their valves the Pectens are so remarkable for, move away into deeper water.

These researches were entirely carried on in the morphological laboratory of the University of Cambridge, and my best thanks are due to Mr. Balfour for his valuable advice and encouragement during the whole course of my researches. Owing to his kindness, also, I have been enabled to examine some of Semper's preparations of the eye of Onchidium, to which reference has been made in the text.

Methods.—For a general examination of the eye the best method is to harden in alcohol and stain by immersion in hæmatoxylin for twenty-four hours. Of the osmic-acid acid preparations the best were obtained by immersion in a 1 per cent. solution for fifteen minutes, followed by absolute alcohol for three or four days. This method is of great value for studying the retina and lens. I have also used gold chloride for staining the nerves with some success. For examining the tapetum the best preparations I have were made from some eyes given me by Mr. Haddon, which had been treated with picric acid. This reagent seems to have dissolved away the red pigment, and consequently left the tapetum free from the numerous little red granules which

generally cling to it. For examining the isolated rods of the retina I have allowed the eyes to remain in a solution of chloral hydrate for four or five days. I have then dissected out the retina with needles as carefully as possible, and poured a drop or two of hæmatoxylin on to the slide. When the retina had been standing in hæmatoxylin in this manner for some hours it was washed with water, teased out with fine needles, and mounted in glycerine.

On the TERMINATIONS *of* NERVES *in the* EPIDERMIS. By
L. RANVIER, Professor at the College de France. With
Plate XXXVI.

IT is well known that by the employment of chloride of
gold it is possible to show the ultimate nerve-endings in
tissues. But this reaction is not always followed with suc-
cess if the original process of Cohnheim is pursued ; and in
attempting, during the last few years, to render it more
regular by combining the action of the gold with formic acid,
Pritchard and Loewit have effected a very notable progress.
At the same time the process of Loewit, although it gives far
more constant results than any previous process, is not
without its disadvantages. The solution of a third part of
formic acid, in which the tissue is placed before being sub-
mitted to the action of the gold, alters the ultimate ramifica-
tions of the nerves, so that, on being fixed by the action of
the gold, they are more or less altered.

It occurred to me that, in the simultaneous action of
formic acid and chloride of gold, the latter reagent would
retain its selective power, and by the rapidity of its action
would prevent the nervous terminations from being seriously
injured by the acid. It appeared to me also important that
the reduction of the gold should be as rapid as possible.
These considerations led me to the following process.

The tissues, with the nerve terminations, are placed in a
mixture of chloride of gold and formic acid, which has been
boiled and then cooled. After remaining in this mixture
between two and four hours they are removed and washed,
and the reduction of the gold is effected either by the action
of daylight in slightly acidified water, or in the dark in a
solution of formic acid.

By this method of treatment the terminations of the
nerves in muscles appear continuously arborescent, instead of
being frequently interrupted, as they are when the process
of Loewit is employed. At the same time they contain some
irregularities which prove that the process is still insufficient.
For this reason it became necessary to invent a fresh process.
I attempted to replace formic acid by an acid which would
not have an equally deleterious effect on delicate elements,
and believe that I have found it in the juice of the lemon.

Lemon juice, though its protracted action alters nervous
tissues, yet preserves their form sufficiently long for it not
to be altered in the time requisite to procure the whole effect

of the chloride of gold. Preparations of the white or red muscles of the rabbit, treated successively with lemon juice and chloride of gold, preserve the nerve terminations, not only continuously arborescent, but also remarkably regular.

I have employed this process in the investigation of the nerve terminations in the epidermis in general, and have employed for this purpose organs in which numerous authors have already studied the terminations of nerves ; for example, the snout of the pig, the nose of the mole, and the skin of the human finger.

In the pig's snout (fig. 1) the nerves which penetrate and ramify in the epidermis have a variable diameter ; the ramifications they form have an equally variable distribution, though on this point it is not necessary to insist. It is, however, to be noticed, as is shown in fig. 1, that in proportion as the nerves approach their termination they become moniliform, and that some of them break up into granules, *which become completely free*. Some of these granules are to be seen completely isolated in the corneous layer of the epidermis.

As regards the termination of the nerves in the nose of the mole (*Talpa europea*), I will assume a knowledge of the investigations of Eimer and Moïsisovicz. I will add that at the base of the epidermic thickening, amongst the nervous tubes destined for them, there are small Paccinian corpuscles, and that in the deeper layer of the epithelial mass forming these papillæ there are five or six small rounded nervous corpuscles, as to the signification of which I do not propose to give any further details. As regard the true inter-epithelial nervous branches (fig. 2), it is important to notice that those which, to the number of two or three, are placed in the centre of the organs of Eimer, form zigzags, which become the more pronounced as they approach the surface of the epidermic covering. At the angles of the zigzags there are at first thickenings of the nervous fibre, and finally veritable buttons. These buttons become more and more conspicuous, become stalked, and in the neighbourhood of the corneous layer become completely free. The marginal nerve-fibres (*vide* fig. 2) of the organ of Eimer remain straight through their whole course, but as they approach the surface swellings appear on them, which are placed in the same transverse line for all the fibres. These swellings enlarge, soon form a prominence turned towards the centre of the organ, then become stalked, and finally become completely free near the corneous layer.

The same process may be observed with the peripheral

fibres of the organ of Eimer (fig. 2). These fibres divide so as to form rudimentary arborescences, which end in buds and button-like thickenings, which can equally be separated, travel towards the surface, and be thrown off to the exterior with the other products of the evolution of the epidermis.

In preparations of the human epidermis (fig. 3) made with chloride of gold, the terminal branches of the nerves have an arrangement analogous to that of the peripheral nervous ramifications of the organ of Eimer. Only the nerves which penetrate into epidermis have a very inconstant diameter, and the ramifications they form are subject to great variations, upon which I need not insist further. I must content myself with saying that in proportion as they approach the corneous layer the ultimate nervous ramifications become more and more moniliform, and finally break up into granules, which are no longer connected with the nervous system, and which, mingled with the epithelial cells, appear, like these latter, to be subjected to a process of elimination.

The theory, or rather the hypothesis, which I propose is founded on the facts which I have just briefly expounded.

The nerves which enter the epidermis, whatever may be the form or extent of their ramifications, are subject to a continuous evolution. They grow while at the same time their terminations undergo a gradual degeneration; this degeneration leads to the formation of granules of nervous substance, which become completely free, and are soon transported into the inert layers of the epidermis.

On the TERMINATION *of the* NERVES *in the* MAMMALIAN
CORNEA.   By E. KLEIN, M.D., F.R.S., Lecturer on
Histology at St. Bartholomew's Medical School.   (With
Plate XXXVII.)

THERE is hardly an organ in which the distribution of
the fine nerves can be so easily observed as in the cornea,
thanks to the invaluable discovery of Cohnheim of staining
the organ with chloride of gold.   Since his publication,
November, 1866, " On the Termination of the Sensory Nerves
in the Cornea," in 'Virchow's Archiv,' vol. xxviii, a very great
number of observations on the same subject have been pub-
lished, all of which have been obtained by Cohnheim's
method of chloride of gold.

On a perusal of all these publications, one arrives at
the remarkable conclusion that, notwithstanding the excel-
lence of the method, notwithstanding the distinctness with
which the final nerves are traceable in all parts of the
cornea, notwithstanding the great transparency and rela-
tively simple structure of this organ, notwithstanding the
absence in it of a variety of structures, such as glands,
blood-vessels, &c., capable of materially interfering with the
observation of the fine nerves—notwithstanding all this, there
exists the greatest variety of opinion as to the termination of
the fine nerves.

The arrangement of the microscopically coarse nerve
branches in the cornea of the frog and mammal were
understood before the use of the gold method, thanks
to the researches of Saemisch, Arnold, and Hoyer; and after
the gold method the observations of Cohnheim, Kölliker,
Hoyer, His, Waldeyer, and others have, one might almost
say, been exhaustive on the same, viz. the arrangement of
the coarser branches; it is the finer and finest fibres whose
termination, nay, even whose distribution and arrangement
is still a matter of discussion.

The last publication on this subject is by Izquierdo,
(' Beiträge zur Kenntniss der Endigung der sensiblen
Nerven,' Inaugural Dissertation, Strassburg, 1879) and by
Waldeyer ('Archiv f. mikrosk. Anatom.,' xvii, p. 367),
chiefly relying on Izquierdo's researches carried out under his
(Waldeyer's) direction.   They are to the effect that the fine
nerves entering the anterior epithelium of the cornea termi-
nate in the superficial layers, in the manner described by

Kölliker and Hoyer, viz. with free ends of the same nature as maintained by Hoyer, while most of those in the substantia propria terminate in the processes of the corneal corpuscles. I shall return to these observations below in detail, but wish to point out here already that they are diametrically opposed to those that I described in this Journal in 1871 (October, 1871), and in the 'Monthly Microscopical Journal' (April, 1872), to the effect, first, that the fine nerves having entered the anterior epithelium and having branched and run horizontally (Kölliker) for a longer or shorter distance, terminate in a network, which I called the intraepithelial network; and secondly, that those in the substantia propria do not anastomose with the processes of the corneal corpuscles, as was mentioned by Kühne, Cohnheim, Moseley, Lipmann, and others, as will be referred to below minutely, but although in their extremely long course many times they come in close contact with the corneal corpuscles, do not terminate *in* them but as a network *on* them. For the reason of this discrepancy between my results and those of Izquierdo and Waldeyer, I have again made the nerves of the cornea a subject of investigation, and I am able to prove that neither do they terminate with free ends in the anterior epithelium, nor are they connected with the corneal corpuscles.

On making this renewed investigation, I have observed several other points which appear to me to be of importance in the discussion of the mode of termination of the fine nerve-fibres.

In order to get at the consideration of the intraepithelial fine nerves and those of the substantia propria, we shall start with the axis-cylinders, which in the front layers of the substantia propria are most numerous, forming what are called by Arnold the subepithelial, by Hoyer the subbasal, and by Waldeyer the fine stroma plexuses. We shall refer to them simply as the stroma plexus. The nature of the branches of this plexus, as bundles of primitive fibrils, the endothelial perineural sheath of the larger branches, the great difference in thickness of the different branches and in the different planes of the same branch, the angular plate-like enlargements, where two or more of them join, are so conspicuous that there are hardly any differences of opinion about them, and it is therefore unnecessary to refer to them more than in passing. There is no difference amongst the recent observers as regards the position of these plexuses. They are all agreed about their being *behind* Bowman's membrane.

First, as to the relation of this stroma plexus to the sub-epithelial nerve expansion.

Cohnheim (l. c.) traced from the most anterior branches of this plexus isolated twigs, which immediately underneath the epithelium split up into bundles of elementary nerve-fibrils. Hoyer (' Archiv f. Anat. and Physiolog.,' 1866, Heft 2) already saw these twigs; and Kölliker (' Ueber d. Nerven Endigung in der Hornhaut Physic. Med. Gesellsch., in Würz-burg,' 30th Juni, 1866), says of them that they pass in an oblique direction through the membrana Descemeti, in order to arrive at the posterior surface of the epithelium, and he therefore called them rami perforantes. Hoyer (' Archiv f. mikrosk. Anat.,' ix, ii Heft, p. 236) was the first who described in the most anterior layers of the substantia propria, but underneath the anterior basement membrane, a special nerve termination, the subbasilar plexus, which consists of fine and finest fibrils derived from the branches of the stroma plexus. Most of the finest fibrils after a long, straight or wavy course and after branching terminate with free ends in the tissue. All fibres of the subbasilar plexus remain underneath Bowman's mem-brane, and this plexus is densest in the peripheral zone of the cornea. While confirming Hoyer's observation as to the great abundance of the fine and finest fibrils in this sub-basilar plexus in the cornea of the guinea-pig and rabbit, I am able to add to his description on some important points.

I will here state once for all what in the following pages I shall speak of thick and thin fibrils; as I have attempted in my former paper (1871 and 1872) so also here I distinguish the nerve branches of the stroma plexus, each of which is in fact only a bundle of fine fibrils, as nerve bundles of the first order; these give off fibres, which are to be considered as a small bunch of two or three primitive fibrils, are the nerves of the second order or the *thick fibres*, and the individual fibrils constituting them are the *fine fibrils* or the fibrils of the third order. The latter are characterised by the smaller and larger varicosities more or less closely placed.

The fibres of this plexus are of various thicknesses be-tween that of a smaller branch of the stroma plexus and the finest fibrils marked by regular varicosities; the thicker fibrils have only a short course, since they soon branch into the finer ones. But these latter are of various types. *a.* Such as take at once a bold straight course, as if they at-tempted to run to the furthest possible regions; in this way they bend off from their course once or twice at a right angle,

but in most cases remain in the same level. *b*. Fibrils which take a long and wavy course, altering their direction and level many times, running repeatedly upwards or downwards in a vertical direction. In sections that are placed horizontally, and comprise the anterior layers of the cornea, these fibrils can be traced in their entire course, but it requires great attention to follow them through all their changes of level.

Now, the nerve expansion that Hoyer called the subbasilar plexus can be separated into the following three systems :—1. A plexus of thicker and finer fibrils, which lie in the same level as the most anterior branches of the stroma plexus, that is, immediately behind, and closely to Bowman's membrane. This system we call the *subbasilar plexus proper*. The thicker fibres of this system branch, and they and their branches run a more or less wavy course. The finest fibrils of this system pass out of this level behind or in front. 2. From the branches of the stroma plexus we trace fine fibrils, which at once pass into the layers behind, and here run for a long distance in straight lines ; they branch repeatedly and anastomose by these branches in rare places with similar fibrils ; they terminate apparently in the corneal substance with free ends. These fibrils constitute the deep subbasilar fibrils. We shall return to them below. As mentioned just before, some of the fine fibrils of the first system leave the level of this latter, and run behind it as straight fibrils identical with those of the second system. 3. From the level of the first system fine fibrils pass anteriorly into the basement membrane ; here they may be followed in their winding course for a long distance, changing their direction repeatedly ; they are not very numerous, and within this membrane may be seen to branch here and there. These fibrils may be called the *intrabasilar* fibrils. They and their branches have a two fold destination : *a*, either they again pass backwards and associate themselves to the deep subbasilar fibrils, or, as is not unfrequently the case, they pass anteriorly through the basement membrane and enter the subepithelial network to be described presently. In the Plate XXXVII, accompanying this paper, I have given an accurate representation of these different fibres in figs. 1—3.

The nerve-fibrils in the immediate proximity of the epithelium form what is known as the subepithelial network ; it is made up of fine and finest varicose fibrils, and they are derived from two sources :—(*a*) from the rami perforantes ; this source

was already known to Cohnheim, Kölliker, and Hoyer; as Cohnheim very beautifully showed, they come off from the rami perforantes in "bundles," except in a small central zone. But I would add here that there exist ' bundles ' also in the central zone, but they are not so conspicuous, because smaller. All fibrils are possessed of varicosities, and those of the same bundle diverge the further away from their ramus perforans; they form triangular groups, the apex of which is directed towards the periphery.

(b) A few fibrils are derived from the intrabasilar fibres, as mentioned above.

As regards the course of the fibrils of the subepithelial network, they all have a linear, but more or less winding course. Their number is everywhere very great, but there exist remarkable differences in this respect in different specimens. Both rabbits' and guinea-pigs' corneæ, prepared with the chloride of gold, with or without subsequent reduction by formic acid, tartaric acid, warmth, &c., show some striking variations. The best results, i. e. the greatest number of these subepithelial fibrils, I have noticed in corneæ prepared, after chloride of gold, with tartaric acid (see my paper in this Journal, October, 1871, p. 408), or with glycerine (in the ' Monthly Micr. Journal,' April, 1872, p. 157).

In successful specimens so obtained the number of fibrils is here astounding; it is so great and dense that we can truly speak of a special nervous layer formed here by fine fibrils. Such specimens are not common; they are, in fact, rare; perhaps only one out of ten will show them to perfection; but, since we cannot assume that this one would in reality differ from the others, we must necesssarily conclude that this is due to a lucky unknown condition in the method of preparation, and I would again insist on this, as I have done in my former papers, viz. that the description of the distribution of the fine nerve-fibrils is probably the correct one, which refers to the most perfect preparations.

In the specimens that we are now having before us, and of which figs. 7 and 8 give a faithful representation of the nerve-fibrils of the subepithelial network, it will be seen that the individual fibrils are of very great length, but there are differences in this respect. In fig 8 I have drawn, very accurately, a great number of the fibrils (by no means all) of the subepithelial network of a portion of the cornea, amounting in length to about 0·4 mm., and it will be found that some fibrils exceed this length, while others fall far short of it.

Two facts are very conspicuous in this figure, viz. first, that most of the fibrils branch and really anastomose with another, and secondly, that, without exception, they leave the subepithelial network sooner or later, according to their smaller or greater length. With regard to the first of these facts, it will be noticed, from figs. 7 and 8, that not only do the fibrils of the same " bundle" anastomose with one another, but also those of neighbouring ones ; that consequently the subepithelial fibrils are connected into real networks, as Cohnheim distinctly stated in his paper, and as I have also described and figured it. Hoyer (l. c., p. 232) agrees with Engelmann that the fibrils cross only, but do not anastomose, and Izquierdo (l. c., p. 26) mentions anastomoses between the fibrils of the same bundle, but is not certain whether this is the case also between the fibrils of neighbouring bundles.

The second fact above mentioned, viz. that all of the subepithelial network pass out of the level of this latter is easily ascertained in the preparation represented in fig. 8. It will be found here that *many of the fibrils possess short lateral branchlets, which, soon after they are given off, curve hook-like, and enter the anterior epithelium.* Some of the fibrils of the subepithelial network are possessed of a great number of these minute side branchlets (in the figure these are marked by a small cross), and when viewed from the surface (see fig. 8) they resemble a sort of fishing line, to which are fastened right and left a series of hooks ; in our case the hooks are the little branchlets ascending into the epithelium. But also the extremity of the chief fibril itself ascends into the epithelium. In accordance with this a vertical section through the cornea show these fibrils of the subepithelial network, creeping along the lower surface of the epithelium, and giving off short branchlets, vertically ascending into the epithelium, between the cells of its deepest layer. I do not find in the description given by Hoyer, Cohnheim, Kölliker, Engelmann, Izquierdo, and others, that these minute side branchlets, as marked by a small cross in fig. 8, have been known to them in the form as they are represented in this figure. Kölliker and Hoyer have pointed out that the intraepithelial network is situated immediately underneath the epithelium, between it and the basement membrane ; Krause and Izquierdo, however, think that its fibrils are sunk in between the extremities of the deepest epithelial cells. I cannot admit this latter view, since I possess preparations in which the whole epithelium, inclusive of the deepest layer, is clearly removed without

disturbing the subepithelial network, which is left untouched. Cohnheim (l. c., p. 41) mentions this already of the cornea which has been macerated in acetic acid after chloride of gold, and I can fully confirm him herein.   This could not possibly be the case if the subepithelial network were not situated *underneath* the deepest layer of the epithelial cells. From vertical sections of the rabbit's cornea we obtain the conviction that the greatest number of these fibrils are situated above (*i.e.* anterior to) Bowman's membrane. Cohnheim does not admit for the rabbit's cornea a structure comparable to the similar membrane in the human cornea ;. there can be no doubt about its presence, and in gold specimens it is occasionally well marked as a conspicuous layer of the thickness of a lamella of the ground substance, from which it differs by its being differently coloured, but it is of the same tint as the membrana Descemeti.

Having passed into the epithelium, the nerve-fibrils ascend in a vertical manner, winding their way between the columnar cells of the deepest layer, as was first seen by Hoyer, and afterwards, by means of his chloride of gold method, minutely traced by Cohnheim.   As regards the fate of these intra-epithelial fibrils there is even less agreement than of that of the subepithelial ones.   With the exception of Inzani, Than-hoffer, and Ditlevsen, all observers agree that the intra-epithelial fibrils extend to near the surface of the epithelium. According to Cohnheim (' Virchow's Archiv,' Band 38), having arrived in the superficial layers, some of them branch, others not, and then they change their vertical course into a horizontal one.   Some of them terminate with a minute end knob freely floating in the precorneal fluid.   Kölliker followed the last termination of the intraepithelial fibrils as horizontal fibrils situated between the most superficial layer of the flattened epithelial cell.   Here they run for a longer or shorter distance, branch repeatedly, and anastomose but rarely, and finally each fibril terminates with an end-knob underneath the most superficial layer of the epithelial scales, Engelmann (' Die Hornhaut,' Leipzig, 1867), whose obser vations refer only to the cornea of the frog, saw only free ends of the intraepithelial fibrils between the epithelial cells of the most superficial layers.   At a similar conclusion arrived also Tolotschinow (Inaugural dissertation (St. Peters-burgh, 1867).   According to Petermöller the intraepithelial fibres form networks in all layers of the epithelium.   In my paper on this subject in this Journal I have described them as giving of lateral branchlets, and as forming a deep and

a superficial intraepithelial network, the former situated in the layer next to the deepest columnar cells, the latter below the most superficial layer. The fibrils of the latter branch repeatedly, and run a horizontal course sometimes for considerable distances; they join, cross each other, and wind their way between the epithelial cells, changing their level several times. I have also stated that the end-knobs that one meets with on some of these fibrils cannot really possess this character, since they are absent in others; and, besides, they are identical with those larger varicosities that occur in the course of most of the fibrils.

Hoyer ('Archiv f. mikr. Anat.,' Band ix, p. 234) describes the intraepithelial fibrils in the same way as I had done, but, while admitting anastomoses of these fibrils, maintains that they terminate with free ends amongst the superficial layers of the epithelial cells. The small and large varicosities which these fibrils show in some specimens are absent in others, and, therefore, must be regarded as produced artificially. Rollett ('Stricker's Handbook,' article "Cornea," p. 1136) does not see any anastomosis amongst the intraepithelial fibrils, but says, that having branched they terminate in free ends, often with a slight enlargement amongst the superficial epithelial layers. Waldeyer (Graefe and Saemisch, 'Augenheilkunde,' p. 210), on the other hand, maintains a terminal network of the intraepithelial nerves. Krause ('Allg. und Mikr. Anatomie,' 1876, p. 539) saw the intraepithelial nerve fibrils forming plexuses near the surface; they terminate with small end-knobs. Izquierdo (l. c., p. 27) finds that the intraepithelial nerves, having ascended in a vertical or oblique direction into the superficial layer, bend off into a horizontal course and branch. They do not anastomose. All fibrils terminate with free ends, or with a slight swelling.

Waldeyer ('Archiv f. mikrosk. Anatom,' Band xvii, p. 379), while accepting Izquierdo's conclusions, admits the incorrectness of his former view of an intraepithelial terminal network. When speaking of a terminal network (Graefe and Saemisch, p. 270), Waldeyer then observed also free ends, but "the possibility always remains that in such apparently free ends there is only an imperfect action of the chloride of gold." This is just what I have urged in my first paper in this Journal, 1871, knowing well the great differences in the number of the intraepithelial nerve-fibrils that one meets in various specimens, and in various places of the same specimen, prepared after the same plan, and seeing that in

many cases the nerve-fibril can be traced beyond an apparent end-knob.

In his last paper ('Archiv f. mikr. Anat.,' xvii), however, Waldeyer thinks (p. 329) that the reason for his formerly maintaining an intraepithelial terminal network lies perhaps in the fact that he had chiefly to deal with the human cornea, in which the cement substance becomes readily stained with the chloride of gold, and hence the appearance of anastomosis is easily accounted for. In the preparations that were the basis of my memoir in 1871, and in those of the present occasion, the intraepithelial nerves are the only fibres stained black with the chloride of gold. The epithelial cells and their interstitial substance is unstained, except a faint greyish tint in the deepest layer. The nuclei of the superficial epithelial cells are just visible, being faintly stained violet. I must repeat that in my descriptions of the intraepithelial nerves *I always had to deal with such specimens only.* Those in which also the interstitial substance of the cells in the superficial layers are distinctly stained have always been discarded as unsuitable, since a confusion of the interstitial cement substance with fine nerve-fibrils is then quite possible. This has been ably urged by Cohnhein (l. c., p. 31), and in the drawings of the intraepithelial nerve-fibrils accompanying his memoir (figs. 7 and 8, plate xii); in the figures given by myself in this Journal October, 1871, figs. 2, 3, and 4, Plate XIX; and in ' Monthly Micro. Journal,' April, 1872, figs. 5 and 6, plate xiv; in those represented by Hoyer ('Archiv f. mikr. Anat.,' ix, fig. 2, plate xiii,) the unmistakable and conspicuous nature and course of these fibrils can at once be recognised. To all those who have seen good specimens of the intraepithelial nerves it must be evident that, judging from the representation given in Rollett's fig. 390, l. c., p. 1136, and in Waldeyer's fig. 21, l. c., p. 210, both these authors had not seen the intraepithelial horizontal nerve-fibrils of the superficial layers, and consequently were not in a position to judge of their final distribution.

Following, then, carefully in a horizontal section—the anterior free surface being directed upwards and a place being selected that comprises only the epithelium—of a well-stained and well-reduced gold cornea, the intraepithelial nerve-fibrils, after they have passed between the columnar cells, can be traced as more or less horizontal fibrils in all layers anterior to the deepest columnar cells. The length of these fibrils varies very greatly; some of them are very short, not longer than the breadth of two or three epithelial cells, while many others may

be traced for a very considerable distance, even through several
fields of the microscope. On their way they give off right and left
similar horizontal fibrils; every one of them, if long enough, can
be seen to change repeatedly its level, now ascending to almost
the free surface, then, again, dipping down and pursuing its
course among the middle layers of the epithelium.   In this
manner they form a very intricate texture, such as I have
figured in my former papers, and as is also figured by
Hoyer in his fig. 2, plate xiii.   We find these fibrils in all
layers, between the most superficial layer of scales and the
deepest layer of columnar cells, and I must, therefore, correct
accordingly my former statement as to the presence of these
fibrils only in two layers (a deep and superficial intra-
epithelial network), although I must adhere to my former
statement that they are most numerous in the superficial
layers.   The fibrils are of various thicknesses ; some are con-
spicuously thicker than those of the subepithelial network.
This is especially the case with those in the superficial layers,
as I must persist in maintaining against Hoyer (l. c., p. 234) ;
but in all layers in which they occur we find fibrils of com-
paratively great thickness crossing and anastomosing or
giving off respectively fibrils of such fineness that it is just
possible to trace them.   Whether very fine or not, they almost
invariably possess minute varicosities, some fewer, others
more ; especially the very fine ones show them better and
more regularly disposed than the thick.   Besides these fine
varicosities there are other larger ones more irregularly
distributed.   They are either staff-shaped, or spherical,
or pear-shaped, or angular.   Any of these large vari-
cosities may be met with in the course of the fibrils
where they join others or where they give off one or two
lateral branches, or they occur at the apparent extremity of a
fibril. I say purposely "apparently," since we shall presently
see that they are not the real ends.   Such apparent ends
occur on the fibrils or their branches in all layers, as Cohn-
heim very correctly described (l. c., p. 28), except, of course,
the deepest layer of columnar cells, and the most superficial
one of flattened scales.

The nerve-fibrils do not pass into this last-named layer, and certainly do
not extend beyond it. I have expressed myself in the same sense already in
my former communications, in accordance with Kölliker. Hoyer and
Izquierdo did not see them pass into the most superficial layer of scales.
When looking at a horizontal section comprising the complete epithelium,
it will at first appear as if some of the most superficial intraepithelial
fibrils did really extend as far as the free surface.   To determine this it is
necessary to use high powers, such as Zeiss's $\frac{1}{12}$ oil immersion, or Hartnack's
10 immersion ; it will then be seen that one layer of nuclei (*i.e.* those of

the most superficial scales) can be traced above the most superficial nerve-fibrils. On account of the great thinness of the superficial scales even a moderately high power, such as Zeiss's D and E ($\frac{1}{6}$ and $\frac{1}{9}$ inch), or Hartnack's 7 and 8, does not suffice. Amongst the great many specimens which I have examined—they amount to hundreds—only in very rare instances have I seen one or the other fibril at the same level as the most superficial nuclei.

Before coming to their ends I must say a few words as regards their relation to one another. Cohnheim (l. c., pp. 27 and 28) does not speak of any anastomosis of the intraepithelial nerve-fibres, while Kölliker (l. c., p. 4) observed such anastomoses, "although, as it appears, not frequently." Anastomoses of the intraepithelial nerve-fibrils, although, on the whole, rarer than I thought them to be in my first memoir, do no doubt occur. My mistake of assuming them to be of very frequent occurrence arose from my not having examined with sufficiently high powers.

It must be borne in mind that these fibrils in many places, when crossing, are in very close contact, and if they happen to cross at the point of a varicosity the appearance of an anastomosis is produced. When examining such places with an oil immersion $\frac{1}{12}$ of Zeiss I can ascertain that in four out of six there is really no anastomosis between the fibrils; in the other two it is impossible to say that there is not an anastomosis. But there exist real anastomoses, about which there can be no doubt whatever; these refer to such places where two of the long fibrils are joined with one another by a shorter or longer side branch. In fig. 10 are represented such undoubted anastomoses.

Now, as to the larger or smaller knob-like structures which are met with on the apparent extremities of the shorter as well as the longer fibrils, as mentioned above, and as observed by all who have worked at this subject, there is no doubt about their presence, and the question is whether they are the real ends or not. Kölliker (l. c., p. 4) would not say that these knobs are natural or artificial, Hoyer (l. c., 234) considers them as artificial products, and Izquierdo (l. c., p. 28) asserts that some fibrils, at any rate, possess them. Cohnheim, Tolotschinow, Krause, and others, consider them as natural end-knobs.

It cannot be denied that they possess the same appearance, whether at the end of a fibril or in its course; but the great differences in their number, size, distribution, and shape, seems to point to their being artificial products. That some of them are not "end-knobs" this I am going to prove presently; that others are such is doubtful, since it cannot with any show of reason be asserted, as Izquierdo does, and

as Waldeyer confirms, that there exist two modes of termina-
tion—one with end-knobs, and the other without.

What is, then, their real termination ?

In preparations which would pass the muster of good
nerve preparations, there is nothing to be seen, even when
examined with a high power, beyond the fibrils as figured
by Cohnheim, myself, Hoyer, and Izquierdo, and as now re-
presented in the figures of the Plate accompanying this present
memoir. But after they have been kept mounted in glycerine
for some time—I refer here to specimens of the rabbit's cornea
prepared after the simple (Cohnheim's) method of chloride
of gold, as well as that where afterwards a reduction is
effected by means of tartaric acid (see my former paper in
this Journal) and mounted in glycerine for one or two years
—and if they are then carefully examined, it will be found
that in some of them a great many exceedingly fine fibrils
are seen, which are given off by the above ordinary intra-
epithelial fibrils, both as lateral branchlets and from the
knobs that previously appeared as end-knobs. These
fibrils, which at the time the preparation was mounted
could not be seen, are now distinctly traceable, some
for a long, others only for a short distance. In some
places they appear as a row of minute stained dots with
unstained joints between, while in others both the dots
and joints are stained and then easily perceived. Fol-
lowing under a high power, e. g. Zeiss' oil immersion $\frac{1}{12}$,
which by its exquisitely fine definition is admirably suited
for this purpose, one of the fibrils in the superficial layers—
of course, only thin horizontal sections of the epithelium
being referred to here—we are at once struck *by the very
great number of short lateral rod-like projections which are
seen on most fibrils, both the ordinary ones as well as the very
fine ones* just mentioned. Generally, but not by any means
always, they are given off at the varicosities. Especially
interesting are, in this respect, the knobs which were above
referred to as end-knobs ; some of these appear beset with
these projections on their whole circumference, and resemble,
then, somewhat the thick extremity of a stamen with its
radicles. Most of them are very short, rod-like, others can
be traced for long distances, and in this case they are in-
variably dotted, and present themselves as regularly varicose
fibrils, giving off likewise numerous lateral branchlets. Also
in this case, either only the varicosities appear stained and the
joints unstained, or the joints are faintly stained. Now, both
the short rod-like as well as the longer filamentous branchlets,
ramifying dendritically and uniting by their branchlets,

break up into a network, which is of exceeding minuteness. The network appears composed of small rods of equal length, and at all the nodal points there is a minute dot or granule. These rods appear generally faintly stained as compared with the dots, but in some places they are stained as much as the latter. Only in few places have I seen this network very perfect, and then only over a small space, but then it appeared with sufficiently great distinctness as a reticulated plate-like expansion of some of the fine nerve-fibrils. This minute network, which I shall call *the terminal network*, is shown in figs. 11, 14, 15, 16, and I must refer the reader to these figures which give a more comprehensive idea of its nature than any lengthy description; it is situated *between* the epithelial cells, since we see it from its broad surface and in profile (see figs. 11 and 16). As mentioned on a previous page, fibrils with end-knobs are met with in almost all layers, and so, I presume, is also the terminal network, but for obvious reasons it is only distinctly traceable in the superficial layer.

In the above figures I have, for the sake of comparison, added a few nuclei of the flattened epithelial cells; these nuclei are just visible as faintly outlined oval or spherical discs, of a pale grey or bluish colour; of the outlines of the cells themselves nothing can be seen.

Thus, it is easily recognised that the nerve-fibrils we have to deal with here as connected with the end reticulum are considerably finer than those drawn by Hoyer (fig. 2, pl. xiii), or Izquierdo (l. c., fig. 5, pl. i). The fineness of the terminal network is by no means inferior to that of the terminal network which I described of the nerves in the skin of the tadpole, and situated immediately underneath the epithelium ('Sitzungsber. d. k. Akadem. der Wiss.,' Vienna, 1870, vol. 61), and also observed by Lavdowsky ('Arch. f. norm. und path. Histolog.,' 1870, and 'Centralblatt f. med. Wiss.,' 1872, No. 17), nor that discovered by Gerlach in the grey matter of the spinal cord ('Centralblatt f. d. medizin. Wiss.,' 1867, Nos. 24 and 25), and by Rindfleisch ('Archiv f. mikr. Anat.,' viii, p. 453) and Gerlach ('Centralbl. f. d. medizin. Wiss.,' 1872, No. 18), in the grey matter of the cortex of the cerebrum. If Waldeyer in his last article ('Archiv. f. mikr. Anatomie,' xvii Band, iii Heft) sums up by saying (p. 379) that there exist no terminal networks, and that the fine nerve-fibres ending in the epithelium, either run out freely or end with a minute knob, it is necessary to remind the reader that Waldeyer bases himself on Izquierdo's specimens, which, as far as refers to the intraepithelial nerve-

fibrils, I consider imperfect, and Waldeyer's assertion must therefore be taken to have the value of negative evidence only.   That the reduction of the gold in Izquierdo's specimens has not been complete is further proved by his saying (l. c., p. 29) that, " in vain have we searched in the epithelium for the cells of Langerhans, which Ribbert pretends to have recently found."   These cells—elongated cells with very many finer and thicker branched processes—it is true, are not met with in ordinary specimens, but in those in which the reduction of the gold has been complete they are very conspicuous by their colouration and size.   Their number generally varies in the middle and superficial layers of the epithelium in different places and their processes, although crossed by the intraepithelial nerve-fibrils, appear nowhere connected with them.

The assertions of Inzani (quoted by Izquierdo, l. c., p. 16) about special terminal ends situated amongst epithelial cells of the deepest layer and possessed of minute fibrils, as well as those of v. Thanhoffer ('Virchow's Archiv,' Band 63, 1875) about the fine nerve-fibres terminating in so-called tactile corpuscles, situated between the cells of the deepest layer of the epithelium, are obviously due to imperfet specimens.   This is well illustrated by the fact that Thanhoffer did not see any nerve-fibrils beyond the deepest layer of cells.   To a similar cause, viz. to imperfect specimens, must be also attributed the assertions of Beale about the absence of nerves in the epithelium.   Beale did not see any nerve-fibres in the anterior epithelium of the cornea, but then this observer did not make use of the chloride-of-gold method.

The last point which I wish to discuss here is the termination of the nerve-fibrils in the substantia propria.   As is well known, Kühne was the first to maintain that in the frog's cornea the finest fibrils terminate in connection with the processes of the corneal corpuscles.   The number of observers who made this a special point of investigation, although great, range themselves in those who confirm Kühne, Moseley (this Journal, July, 1871), Königstein ('Sitzungsber. d. K. Akad. in Wien.,' 1877, Band 76), Izquierdo (l. c., p. 25). and Waldeyer (l. c., p. 378) ; both these last-named observers assert, however, that some fine fibrils terminate also free in the substantia propria.   Then those who simply deny such a connection, and accept a free ending of the fine nerve-fibrils in the substantia propria, Kölliker (l. c.), Engelmann (l. c.), Dwight (' Monthly Micr. Journal,' 1869),

Tolotschinow (l. c.), Rollett (l. c., p. 1138)—who is, however, not able to say in what way they terminate—Hoyer, and Krause (l. c.). And, lastly, those who maintain a special termination of the fine nerve-fibrils, Lipmann ('Virchow's Archiv,' Band 78, p. 218), who says that the fine fibrils terminate in the nucleoli of the nuclei of the corneal corpuscles. Lavdowsky ('Archiv f. Mikr. Anat.,' viii) assumes two kinds of terminations of the nerve-fibrils, (a) in rhombic plates, supposed to be present in the wall (?) of the lymph-canalicular system (dog), and (b) with peculiar enlargements near the nucleus or in the nucleolus (frog, dog, cat, calf).   In my former papers I have stated that the finest nerve-fibrils in the substantia propria, in good gold specimens of the frog's cornea, are conspicuous by their great length, their peculiar rectilinear course, their right-angled bending, their few branchings ; these fibrils I described as the nerves of the third order, and as being nowhere in connection with the corneal corpuscles, although they come in close contact with them in many places ; they are not the finest nerve-fibrils, as assumed, since in some places they are seen to give off very minute short fibrils—nerve-fibrils of the fourth order—which, on the surface of the corneal corpuscles, anastomose in a sort of network.

I have now extended my observations on the fine nerve-fibrils in the substantia propria of the cornea of the kitten and rabbit, and I must also for these maintain that the fine nerve-fibrils that one sees in ordinary gold specimens are not the finest and last fibrils.   Especially easily followed are the fine nerve-fibrils in the anterior strata of the substantia propria, where they form what we considered above as the deep subbasilar fibrils, and which have been very beautifully described already by Hoyer.   But also in the deeper sections of the substantia propria similar fine nerve-fibrils can be met with.   Their general character is that they may be traced for very long distances ; that they possess now a rectilinear, then, again, a wavy course ; that they keep close to the corneal corpuscles, always running in the lymph-canalicular system, as was already known to v. Recklinghausen, and as was very extensively described by Lavdowski and Hoyer ; that they give off from place to place a lateral branchlet ; that in some parts they are seen to bend off suddenly at a right angle, either after a long rectilinear or curved course, or at short intervals ; further it will be noticed that they continually change their level, now passing into a stratum above or below, then again returning into their original plane; and,

finally, that they anastomose, in a few instances, with their neighbours.

These fibrils differ in thickness, and they all contain small or large varicosities, more or less regularly disposed. I must draw the attention of the reader to figs. 4 and 4A, and 5, in which the nature and course of these fibres is drawn very accurately. In what way do they terminate? As is well shown in the figures, after having given off one or more branches, they, as well as their branches, appear to terminate freely either at a varicosity or beyond it, at the side of a corneal corpuscle. In this respect I can add little to the description given by Hoyer of these fibrils with admirable faithfulness.

In the cornea of the frog these fibrils are much more diffi- cult to trace, since their great number is compressed into the posterior strata of the substantia propria, that is, into a very limited space. For this reason we find that anastomoses between neighbouring fibrils are more common than in the cornea of the rabbit and kitten.

I possess specimens both of the cornea of the rabbit and kitten, and also of that of the frog, wherein I find places in which these fibrils, when examined with a high power (Zeiss' F, or, still better, oil immersion $\frac{1}{12}$th), give off very minute short fibrils, which close to the surface of the body of the corneal corpuscles give off short, exceedingly fine, dotted fibrils, which themselves are connected into a network. It is, of course, easily understood, that in many instances it is impossible to distinguish between dots that are contained in the substance of the corneal corpuscles and the dots that mark these ter- minal fibrils; but in certain other instances this distinction is possible, viz. in those instances in which *the corneal cor- puscles and their processes are stained only a greyish tint, while the nerve-fibrils and their varicosities, owing to the complete reduction of the gold salt, possess an almost black colour.*

In some specimens, however, we find also nerve-fibrils in connection with the processes of a corneal corpuscle, such as is described by Moseley, Königstein, and Izquierdo, and there seems no mistake about the nerve-fibrils being here directly continuous with a corneal corpuscle. But let us for a moment inquire, What are these specimens? As is known to everybody who has examined a number of corneæ, either prepared after the simple (Cohnheim) method of chloride of gold, or, in addition to this, after using various reducing agencies, such as oxalic acid, formic acid, methylated spirit, or simple heat, hardly a single cornea is obtained in which the sub- stantia propria appears throughout of the same tint. In

some corneæ in a greater or smaller portion, the corneal cor-
puscles and their processes are of a deep reddish-purple or
reddish-black colour, while in others the corneal corpuscles
are hardly at all stained, or only of a light grey or bluish-
grey colour.   The same difference may occur in one and the
same cornea, and, of course, we then find zones of transition
of the former into the latter.   Let us take a cornea in
which the corneal corpuscles and their processes are con-
spicuously brought out, and well and deeply stained, and let
us add that in the majority of these instances also the fine
nerves are well stained, i. e. of the same deep colour as the
corpuscles and their processes.   And let us bear in mind
that the nerve-fibres are situated in the lymph-canalicular
system which also contains the corneal corpuscles and their
processes.   Let us further bear in mind that these nerve-
fibrils and their branchlets run out close to the corpuscles
and their processes.   Under these circumstances nobody will,
I think, be in a difficulty to explain the above-mentioned
connection of the nerve-fibrils with the corneal corpuscles.
Supposing in the preparations represented in fig. 4, 4A, 5,
and 6, the corneal corpuscles were as deeply stained and of
the same colour as the nerve-fibrils, nobody could fail to find
here connections between the former and the latter ; and,
indeed, in the very same specimens from which these draw-
ings were made there are places in which, owing to the deep
colouration of the corneal corpuscles and their processes, the
distinction between them and the fine nerve-fibres is lost,
and, therefore, an apparent connection between them is
found to exist.   It is quite clear that such places are useless
for determining the relation between the nerve-fibrils and
the corneal corpuscles, and it is to me inexplicable how
Izquierdo omitted to bear this in mind, since Hoyer (l. c.,
p. 241) had already drawn attention to the great importance
of having specimens in which the corneal corpuscles and
nerve-fibrils are of a different colouration.   Izquierdo's
drawings (plate i, figs. 1, 2, 3, and 4), leave no doubt that
he had to deal with specimens in which the corneal cor-
puscles and their processes and the nerve-fibrils were very
deeply and uniformly stained.   For this reason, then, his
assertion about the direct connection of the processes of the
corneal corpuscles with the fine nerve-fibrils, and conse-
quently also Waldeyer's summing up (l. c., p. 378) based
upon it, loses the value attributed to it.

HISTOLOGICAL NOTES.   By E. KLEIN, M.D., F.R.S.

I.—*Ciliated Epithelium of the Œsophagus.*

E. Neumann ('Archiv f. mikr. Anat.,' vol. xii, p. 570) observed on the surface of the mucosa of the œsophagus of the human fœtus, between the eighteenth and thirty-second week, ciliated columnar epithelium. This did not, however, form a continuous lining, since in many places the epithelium was stratified pavement epithelium, such as is found in the new born and adult. Kölliker ('Entwickelungsgeschichte, d. Menschen,' &c., 1879, vol. ii, p. 853), found also ciliated columnar epithelium in various places in the œsophagus of the human fœtus between fourteen weeks and the sixth month.

Mr. Anderson and Mr. Ricketts, students at St. Bartholomew's Medical School, when examining sections of the cervical portions of the (hardened) œsophagus of a newborn child—full time—found, amongst the stratified pavement epithelium of the ordinary description, places where the superficial cells were not flattened but columnar; underneath these were polyhedral cells in two or three layers, and finally, a deepest layer of columnar cells. The superficial cells were either without or with cilia; the latter were short and very delicate. The ciliated cells were seen, more or less continuous, chiefly in the grooves between the folds of the mucosa; in the neighbourhood of these there were generally only isolated ciliated cells.

II.—*Cilia in the Central Canal of the Embryo Chick.*

Examining with Mr. Anderson the fresh embryo of a chick of about the stage figured in Kölliker's 'Entwickelungsgeschichte,' p. 138, as of the end of the second day, with seventeen protovertebræ, in warm saline solution and from the surface, I saw in the whole extent of the protovertebræ the central canal of the central nervous system lined with very beautiful delicate cilia; their length was about half that of the cells lining the canal, and they were in very active movement. It was, in fact, this movement by which their presence made itself conspicuous. It could not be seen in the caudal part. In a second embryo of about the same early stage, observed under similar conditions, the movement was seen distinctly in the commencement of the cervical region, but was soon lost, owing to the wall of the

central nervous system approaching too close from side to side, and consequently the central canal becoming altogether lost. And this state, viz. the shutting-up of the central canal, forms one of the most troublesome impediments to see the cilia and their movement. In embryos of a later stage the examination proves unsuccessful, partly owing to this collapse of the central canal, but chiefly to the relative opacity of the tissue, an opacity far too great to allow the very delicate cilia to be perceived. In embryos of an earlier period, such as possess only about 6—10 protovertebræ, I could not detect either the ciliary movement or the cilia themselves, although in several instances the central canal had not collapsed.

In the central canal of the earlier embryos, as well as in those of the later ones, there were always to be met with a few spherical or oval, coarsely granular, and therefore opaque, corpuscles, either isolated or two or three together; they varied in size, and a nucleus could not be seen in them.

In transverse sections through the above hardened embryos nothing of the cilia could be seen, but this will not surprise since the cilia of less delicate structures are lost during hardening; one of the best known examples is the mucous membrane of the human uterus, and of that of mammals.

III.—*The Glands of the Nasal Cavity of the Guinea-pig.*

As is now well known, the glands occurring in the mucous membrane of the nasal cavity are mucous and serous glands (Heidenhain). The latter form, in the thick portions of the non-olfactory Schneiderian membrane, huge masses, in which the alveoli—convoluted tubes, with lateral and terminal shorter and longer branches—are grouped into lobules, separated, as in other cases, by vascular connective-tissue septa.

The alveoli of the mucous glands present themselves in two different forms: either small, with a small lumen and lined with polyhedral or short columnar cells, each with a spherical nucleus; or large, with a large lumen, and lined with columnar cells of the nature and aspect of mucous cells. There are alveoli in which the two kinds of cells occur side by side. Thus, they resemble the mucous glands of the larynx known through Heidenhain, Tarchetti, myself, and others. The ducts are lined with a single layer of thin columnar cells, as in other mucous glands. At the commencement of the nasal mucous membrane, *i. e.* near the

external opening, where the epithelium of the surface is stratified pavement epithelium, with a superficial stratum like the stratum corneum of the epidermis, also the mouth of the duct of the mucous gland receives a continuation of this latter stratum.   Like the epidermis it stains dark in osmic acid.

The intralobular ducts of the serous glands are identical with the salivary tubes of Pflüger, viz. they are lined with a layer of columnar cells, the outer portion of which is distinctly longitudinally striated.   The intralobular ducts ultimately branch into several shorter or longer tubes, which are very narrow and lined with a layer of flattened cells, and they are therefore identical with the intermediate portions of the ducts in the salivary glands and the pancreas,

The epithelial cells lining the alveoli are columnar, and present themselves in two different aspects according to the state of secretion.   In one aspect, viz. during secretion, they are more transparent, thicker, and appear therefore less columnar.   Their nucleus is spherical or only slightly flattened, and pressed against the membrana propria.   In the other state they are less transparent, thinner, more columnar, and their nucleus is spherical, and not quite so close to the membrana propria; but in both conditions the cell substance is a reticulum, more distinctly so, on account of the greater size of its meshes, in the former than in the latter.

There exists, consequently, also in this respect a complete analogy between these and other serous glands.

In those parts where the mucosa is of great thickness, covered with ciliated columnar epithelium, and containing s maller or larger amount of diffuse adenoid tissue, these glands form also larger groups, situated in the deep submucous section.

In preparations that had been prepared, when perfectly fresh, with a mixture of chromic acid and osmic acid, according to the method of Flesch ('Arch. f. mikr. Anat., Bd. xvi, p. 300), the alveoli of the serous glands, especially those nearest to the surface, appear much larger, their cells being distended by the presence of smaller or larger fat-globules, isolated or in groups; consequently there exists, in this respect, a certain resemblance of these glands and the sebaceous and Meibomian glands. From this it appears, then, probable that the secretion of them is not precisely the same as in the other serous glands; e.g. those at the root of the tongue or the salivary glands.

In the interlobular connective tissue isolated lymph-cor-

puscles may be met with, similar to what has been noticed by Boll and Lavdowsky in the salivary glands.

Fine threads, some with nuclei, some without, can be traced in connection apparently with the cells of the intra-lobular ducts and with those of the alveoli. But whether they are nerve-fibres, as seems probable, or not, could not be definitely ascertained.

An interesting fact, observed here in connection with the large serous gland, is the presence of thin, unstriped muscle cells running in all directions, and forming a plexus, in whose meshes the gland-alveoli are situated. In some places, e. g. the conchæ, there exists here also a plexus of large veins, and the above muscle-bundles form the stroma, as it were, for them, and thus a certain resemblance with a cavernous tissue is produced. This muscular tissue is, no doubt, of some service for the discharge of the secretion, seeing their peculiar intimate relation to the gland-alveoli. In the relatively rigid walls of the nasal cavity such inter-stitial muscle-tissue must be of special value to the glands.

REVIEWS.

Atlas of Histology. By E. KLEIN, M.D., F.R.S., and E. NOBLE SMITH, L.R.C.P., M.R.C.S. Smith, Elder & Co., 15, Waterloo Place.

WE have great pleasure in drawing attention to this very handsome work, which is certainly the most richly illustrated treatise on Histology which has ever been published. It occurred to Dr. Klein that by means of the colour-printing, which has now been brought to so high a condition of perfection, it would be possible to produce illustrations which should be practically fac-similes of the delicately stained sections which histological students make use of for demonstration and investigation. Accordingly Mr. Noble Smith has executed drawings from the choicest preparations made by Dr. Klein, which in colour and form present to the eye precisely the same effect as the preparations themselves when accurately focussed.

The text accompanying the drawings is by no means a mere explanation of the latter, but forms an original treatise on Histology, in which Dr. Klein's views are clearly stated in reference to many debatable questions. The medical student will also find in this work a surer guide to the knowledge of modern Histology than in the older books, even those of high authority and reputation, which, on account of their antiquity, are too often marred by erroneous theory and insufficiency of observation.

Freshwater Rhizopods of North America. By JOSEPH LEIDY, M.D., Professor of Anatomy in the University of Pennsylvania, and of Natural History in Swarthmore College, Pennsylvania.

IT is, perhaps, somewhat astonishing that a treatise on living microscopic organisms should be published, as this work is, under the auspices of the " Geological and

Geographical Survey of the United States Territories." At the same time, the authorities can only be congratulated on the wise liberality which has led them to undertake the production of so valuable a work. We have here figured and described by Dr. Leidy, in the most ample manner, a large series of those freshwater forms which Mr. Archer, of Dublin, was the first to detect, and to study in a special manner. Focke, Franz Schulze, Lesser, Hertwig, and Dr. Leidy, have added to and expanded the rich store, which was first tapped and offered to the zoological connoisseur in the pages of this Journal. To the old and well-known genera of freshwater Rhizopods—Amœba, Gromia, Arcella, Difflugia, Actinophrys—a host of remarkable allies have now been added. In the present fine monograph Dr. Leidy describes many already known genera and species which he has detected on the North American continent, but he also describes several new and very interesting forms, *e.g.* Dinamœba, Ouramœba, and Biomyxa. The volume is illustrated by forty-eight quarto plates, and must be carefully studied by every student of Rhizopod faunæ. We should have been glad had it come into the plan of Dr. Leidy's studies to devote more attention to the determination of minute structure as revealed by powers higher than one of 250 diameters.

A History of the British Marine Polyzoa. By THOMAS HINCKS, B.A., F.R.S. Van Voorst, London.

THE well-known and highly valued series of works on Natural History, published by Mr. Van Voorst, is worthily extended by the two admirable volumes, one of text, one of plates, treating of the British Polyzoa, by the Rev. T. Hincks. In an introduction of a hundred and forty pages, illustrated with numerous cuts, the comparative anatomy and general morphology of the Polyzoa are very fully and clearly discussed—the most recent investigations—French, German, and Swedish, all receiving due notice. In the systematic part which follows the groups of Polyzoa are taken up one by one and systematically characterised, until the whole hierarchy, down to families, genera, species, and varieties, is exhausted. The characteristics of species, as well as of genera, and even of suborders, are necessarily chiefly, if not entirely, drawn from the structure of the hard parts. These are very extensively and beautifully figured in the atlas, so that by the aid of this work the student of

marine zoology will have no difficulty in referring such Polyzoa as he may dredge on the British coast to their proper titles. Naturalists are already deeply indebted to Mr. Hincks for his beautiful volumes (in the same series) on the British Hydroid Polyps. In the present work he has dealt in a masterly way with a still more difficult group. We hope that other similar works may ere long be added to Mr. Van Voorst's celebrated series.

Medusæ and Hydroid Polyps living in Fresh Water.—From experiments made by Mr. Romanes on Limnocodium, and published in 'Nature' of June 24th it appears that the fresh-water Medusa is very intolerant of sea water. It cannot be re-introduced with impunity to the medium in which its ancestors originally lived, and this would seem to imply that a very great length of time has elapsed since the adaptation to fresh water took place. Curiously enough, Mr. Romanes has found that marine Medusæ are not so injuriously affected by brine as the Limnocodium is by sea water. The fact, however, is less astonishing when we remember that the percentage of saline matter in solution in sea water is many hundred times what it is in average pond water, whilst the strongest brine has not a percentage of saline matter many times in excess of that of sea water.

On the whole the tolerance by marine animals of fresh water is a much more frequently observed fact in all classes than the tolerance of sea water by lacustrine or fluviatile forms. It is undeniable that existing fresh water forms have been developed by adaptation from marine forms, whilst it is difficult to cite any instance in which adaptation in the opposite direction appears to have taken place; some few marine Oligochætous Chætopods and Pulmonate Gasteropods being perhaps such instances.

The tolerance by Medusæ belonging to marine species of fresh-water under natural conditions was observed by Mr. Moseley, at Browera Creek, in New South Wales, and from Professor Alexander Agassiz I have received some interesting notes recording similar observations.

Mr. Moseley says ('Naturalist on the Challenger,' p. 272), " After heavy rain the surface water in all the upper parts of the creek is so diluted by the torrent of fresh water from the stream, that it becomes almost fresh ; indeed, at the time of our visit, it was for three or four miles down so little brackish as to be drinkable. At a short depth no doubt the

water was salt. Here are the most favorable conditions
possible for turning marine animals into fresh water animals;
in fact, the change of mode of life presents no difficulty.
Below no doubt the water is always salt, but the fish find a
fluid less and less salt as they rise to the surface. We
caught the mullets in the almost fresh water with a net, and
with them the mussels and crabs. I even saw an abundance
of Medusæ, and a species of Rhizophora swimming in the
creek above the sand-flats, where there was scarcely any
salt at all in the water, yet evidently in most perfect
health."

Professor Agassiz writes, " It strikes me as if the conse-
quences resulting from the finding of this freshwater Medusa
had been somewhat overdrawn. In the first place, we have
two genuine freshwater Hydroids, Hydra and Cordylophora,
and in the second place, as far as my experience goes, it is
not conclusive of so fatal an action of fresh water on Medusæ
as Romanes would lead us to believe in. We have quite an
estuary leading out back of Boston Harbour, extending on
the one side to form what we call the back bay, and beyond
this up the Charles River as far as Watertown, where there is
a dam, about seven miles from the inner extremity of the
harbour proper. Here the Charles River falls into this
estuary as a freshwater stream sufficiently large at times to
affect the saltness of the estuary below it at low tide, so
that at Cambridge, half way from Watertown to Boston, the
water is salt only at the highest tides, quite brackish during
the first half of the ebb, and comparatively fresh during the
last part of the tide. At W. Boston Bridge, about one mile
from the head of the harbour, the water at the last part of
the tide is fresh enough and tastes but little salt. At this
bridge there is an abundance of Hydroids which thrive
remarkably well on the drainage of the district, and grow to an
unusually large size. The species found there which has no
free Medusa is *Laomedea gigantea*. While of the Hydroids
which have free Medusæ we find *Eucope diaphana, Eucope
pyriformis*, and *Obelia commissuralis*. All these species are,
therefore, twice during twenty-four hours exposed to salt
water and to nearly fresh water, and thrive remarkably well
under the treatment, as must of course their free Medusæ,
which I have caught both at high tide and low water—in
salt and in nearly fresh water.

" Other of our Medusæ also find their way into this
estuary, and I have found in fresh water at *low tide* active
Sarsiæ, Tiaropsis, and also Aureliæ, which seemed unaf-
fected by the large quantity of fresh water in which they

were found. Cyanea I have never seen in this estuary
except at high tide. I may mention that the scyphostoma
and strobila of Aurelia is found with the above-mentioned
Hydroids attached to the piles of W. Boston Bridge; but
the scyphostoma of Cyanea I have never found."—E. RAY
LANKESTER.

On the Respiration of the Crustacea.—In the note pub-
lished in the April number of this Journal, a prediction was
hazarded that the Zoea larva of the higher Crustacea would
on examination prove to breathe in the same way as the
Copepoda. Zoeas of Cancer, and probably of some species
of Prawn, received from Mr. T. Bolton, have confirmed this
amply. The respiratory diastole and systole of the rectum
with rhythmical openings of the anus are thoroughly well
marked. It may here be noted that in carmine stainings of
the entire *Copepoda* the stain does not diffuse through the
integument, but up through the rectum in the first instance.
The power of dialysis through the chitinised integument is
slight if at all existent. Now that another place is found
for the respiratory function, it may be denied to the
expanded pleura of the carapace.

This constancy of function in the anus is remarkable, and
indicates that the gills which characterise so many of the
higher Crustacea are secondary formations, long posterior to
the differentiation of the class. What, then, is their origin?
Probably they are in all cases modifications of those pro-
cesses of the appendages which primitively bring about
nutritive currents. In *Daphnia* we have the stage where
these processes are the exclusive food bringers, while they
share respiratory functions with the rhythmically contrac-
tile rectum. And as a parallel to the direction of the bran-
chial current from behind forwards in the Crustacea, we
may cite the Lamellibranchs, where the gills, probably in
origin also parts of the locomotive apparatus (according
to Lankester's view) play an equal part in nutrition and
hæmatosis. To explain body gills we have to invoke either
the principle of the similar modification of neighbouring
parts, or else that of displacement upwards from the appen-
dages. As far as I know, for there are none of the memoirs
on this subject within reach, the development of the body-
gills of the crayfish, which might give a clue, has not been
worked out.—MARCUS HARTOG.
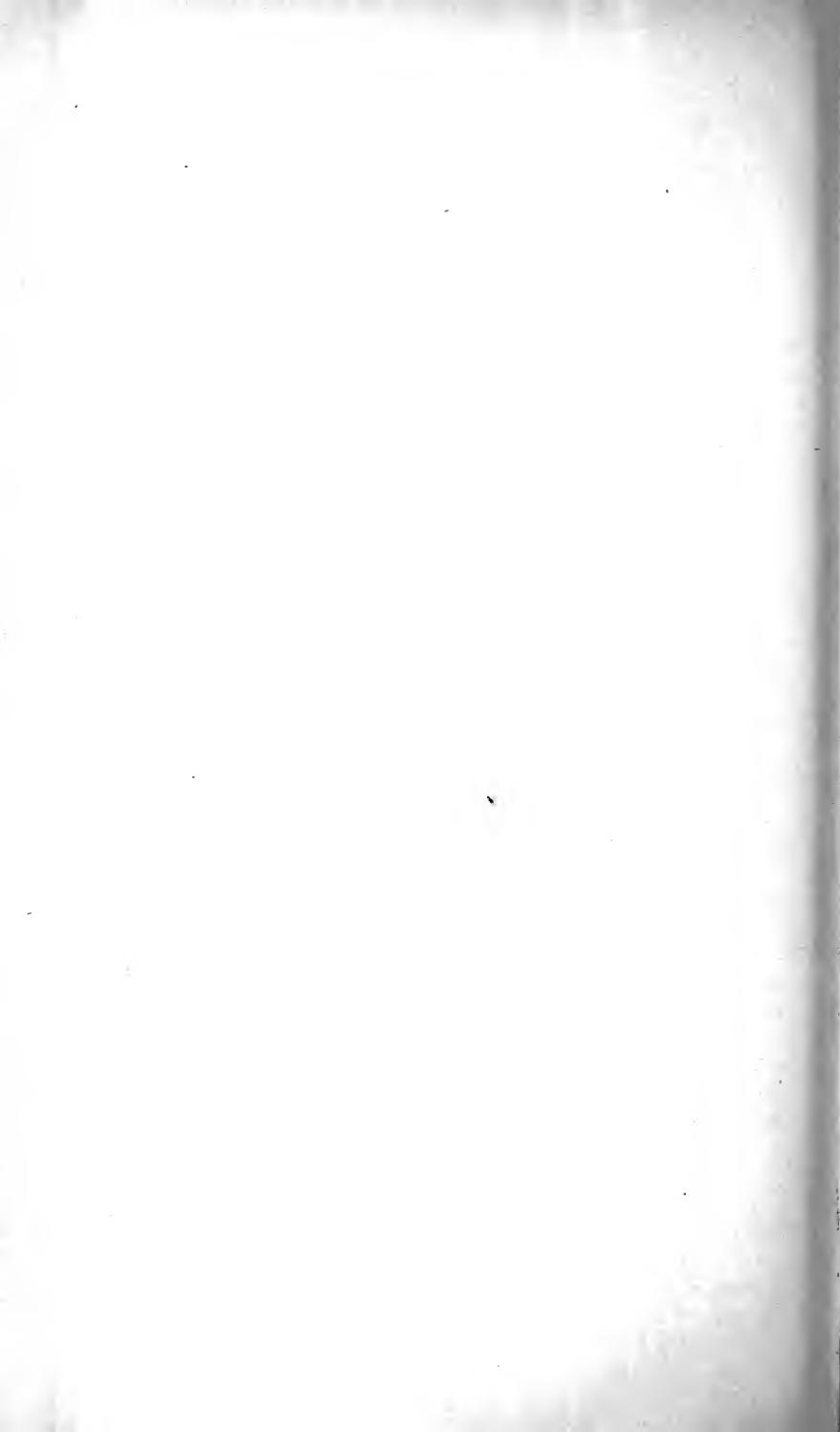
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Fig. 1.

Fig. 2.

Fig. 6.

Fig. 7.

Fig. 14.
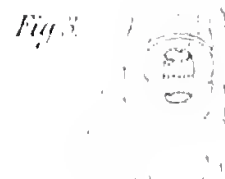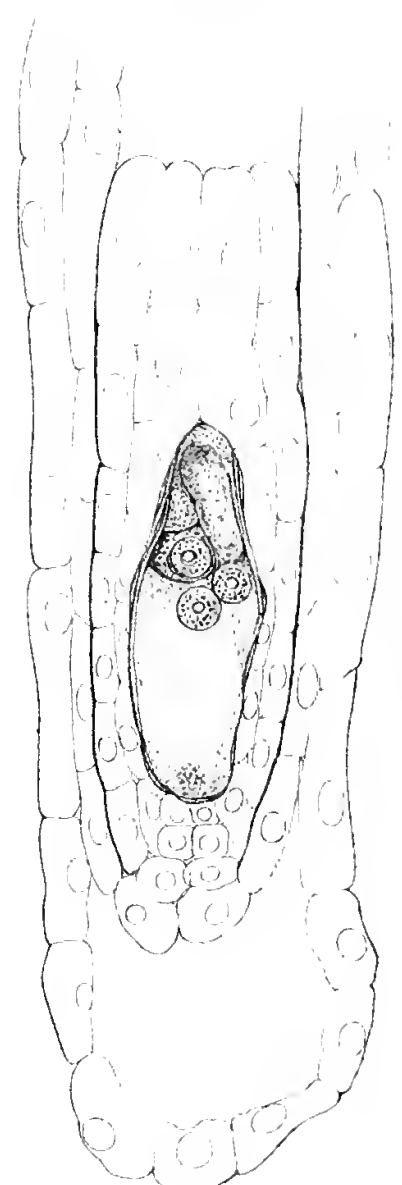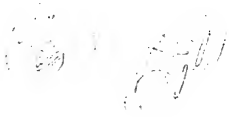
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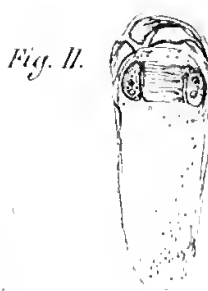
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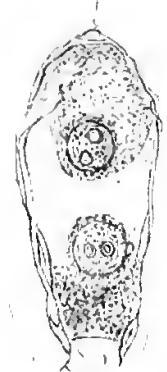
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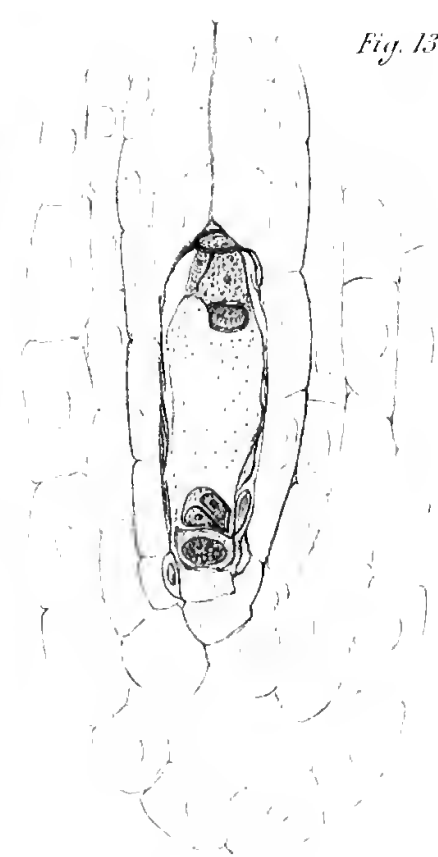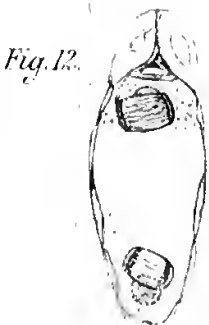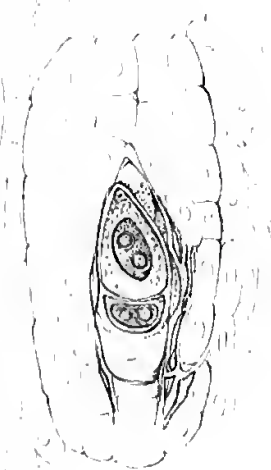
Fig. 12.

Fig. 9.
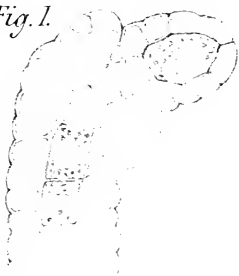
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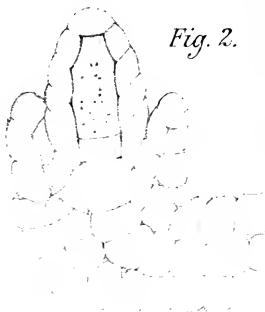
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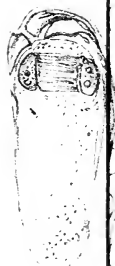
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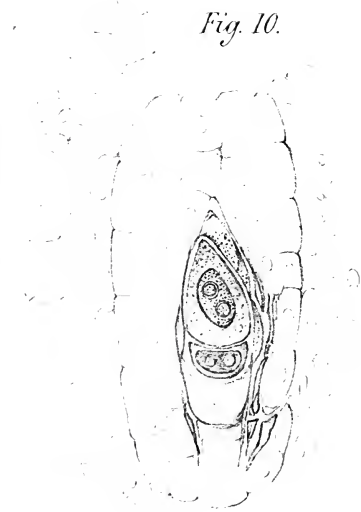
Fig. 12.

EXPLANATION OF PLATES I, II, & III,

Illustrating Mr. H. Marshall Ward's Paper on the "Embryo Sac and Development of *Gymnadenia conopsea*."

N. B.—Except when otherwise stated, the figures are generally from sections in absolute alcohol and glycerine, examined under Zeiss's F, oc. 2 and 3.

PLATE I.

Fig. 1.—Optical longitudinal section of young ovule, showing embryosac mother-cell, and axial row of similar cells. This specimen and the next were examined in the fresh state with traces of ammonic hydrate.

Fig. 2.—Similar section of slightly older ovule, of which the integument is more advanced, and the curvature more decided; the granular contents of embryo-sac mother-cell resist alkalis more than do the others.

Fig. 3.—Actual section through older ovule seen from before. The second integument is well established, and following on the first. The embryo-sac mother-cell has had one "cap-cell" cut off, and a second division is taking place to form the other.

Figs. 4, 5, and 6.—Longitudinal section through the embryo-sac. It is surmounted by two "cap cells," which, together with the surrounding layer of nucleus-cells, are gradually compressed by the enlarging sac. (Fig. 6 is somewhat oblique, but still shows cells of the axial row.)

Fig. 7.—Similar section. Two large nuclei in the embryo-sac have resulted from division. Note traces of cap-cells, and of cells of the nucleus of the ovule around.

Figs. 8, 9, and 10.—Similar preparations; showing divisions of the two nuclei of Fig. 7. Treated (alcohol specimens) with strong acetic acid and fuchsine.

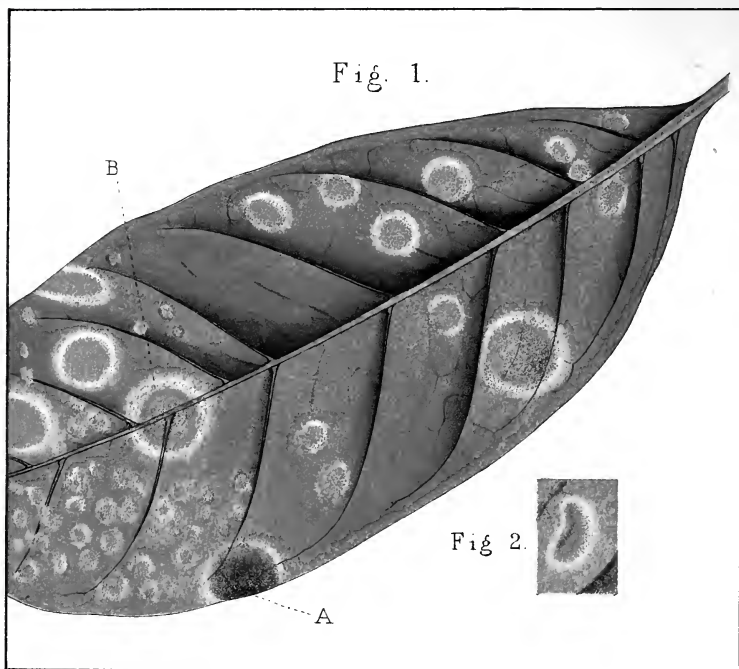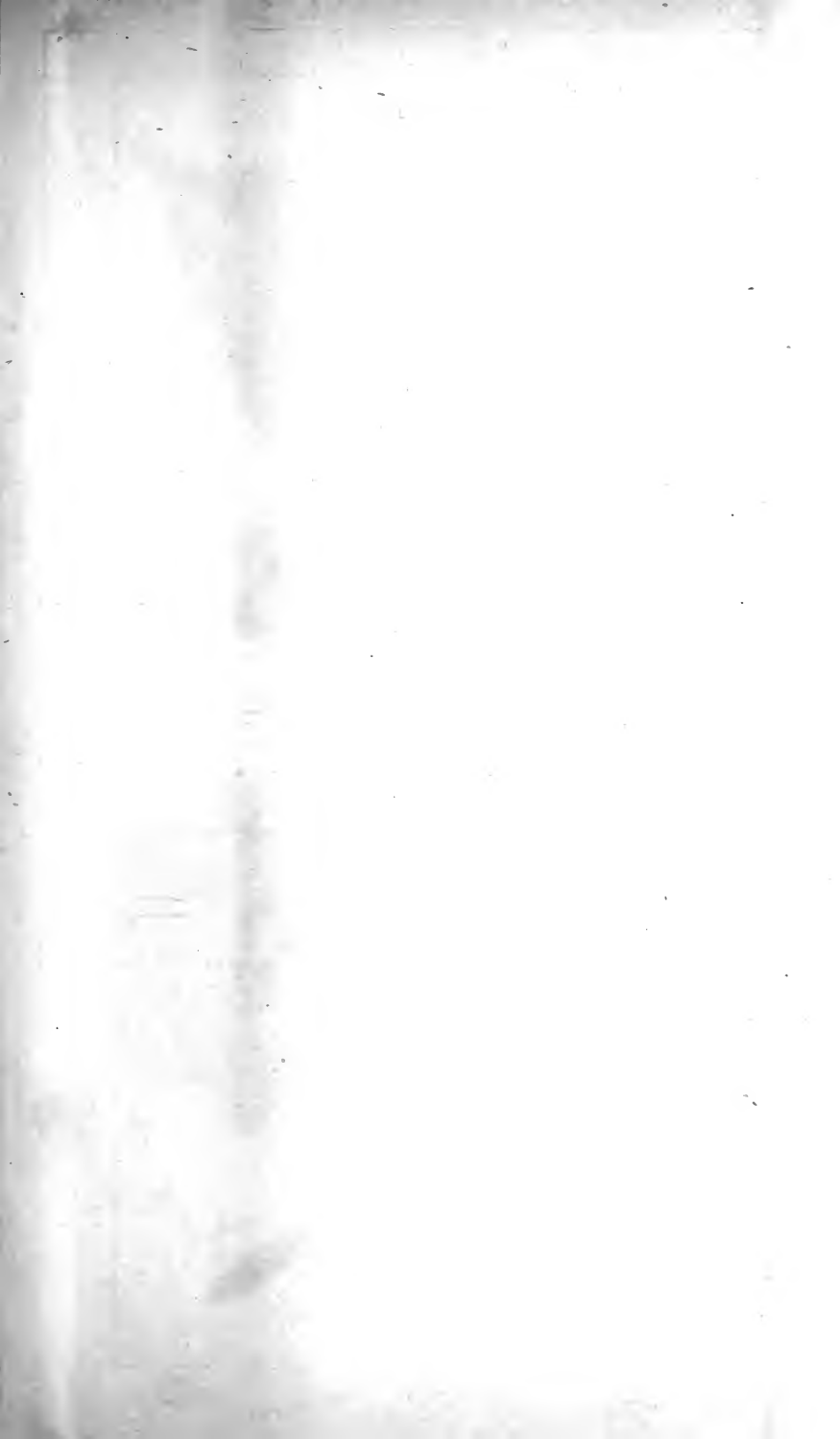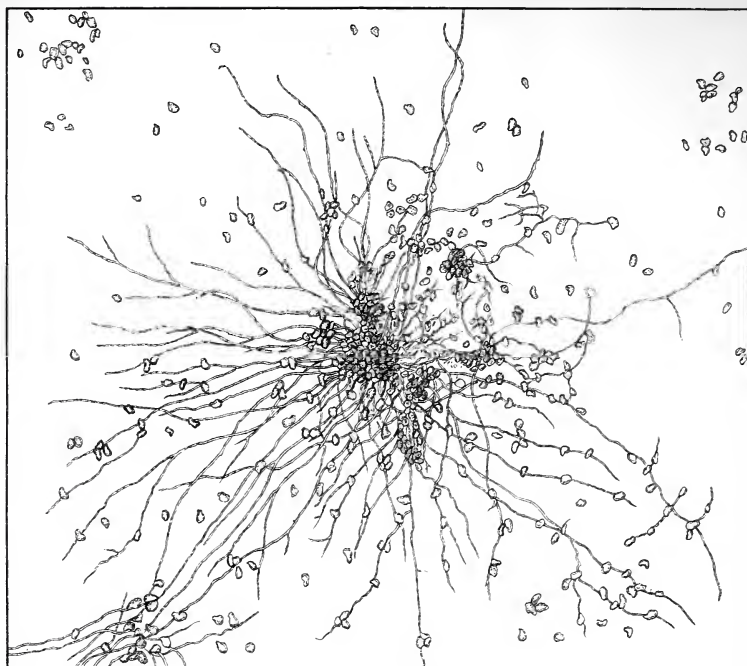Fig. 11.—Similar section, showing division of upper nucleus. Two nucleoli in each half. The section is slightly oblique.

Fig. 12.—Similar section. The two nuclei at each end, and fixed in the act of dividing in planes crossing at right angles; a large vacuole occupies the centre of the sac. Remains of the cap-cells as a refractive mass under the micropyle.

Fig. 13.—Longitudinal section through the ovule. There are four naked masses of protoplasm at each end of the sac. Note traces of cap- and nucleus-ces around. (Alcohol specimen treated with acetic acid and fuchsine.)

Fig. 14.—The masses of protoplasm have arranged themselves in the upper part of the sac as an "egg-apparatus," consisting of two bolster-shaped "synergidæ" supporting an "oosphere" (germinal or embryonic vesicle), and a free nucleus, near which is a second.

PLATE II.

Figs. 15 and 16.—Longitudinal sections through embryo-sac, showing the "egg-apparatus" above, and a free nucleus below, which has travelled down towards the "antipodal" mass.

Fig. 17.—Longitudinal section through a whole ovule during fertilisation. A pollen tube has penetrated the micropyle, and spread its end on the "synergidæ," below and to the side of which is the "egg cell;" at the opposite end of embryo-sac are the "antipodal" cells imperfectly divided, and abutting on these the wandering nucleus from above.

Fig. 17a.—Similar preparation with egg-cell just fertilised.

Fig. 18.—The first division across the fertilised "egg cell" is completed ; remains of egg apparatus and cap-cells are seen above, and of "antipodals" below in the sac.

Fig. 19.—A division appears across each of the two cells in Fig. 18 ; that in the upper cell is not completed. Traces of cap-cells and egg apparatus above, and of antipodal cells below, as before.

Fig. 20.—The embryo now consists of four cells—a "suspensor" of two cells separated by swollen walls, and an embryo proper of two super-posed cells, of which each nucleus contains two nucleoli.

Figs. 21, 22, and 23.—Embryos a little more advanced. The embryo proper becoming divided by vertical walls at right anges, comes to con-sist of three cells ; the lowermost in fig. 23 is dividing.

Fig. 24.—The embryo proper consists of four cells, and the left hand lower nucleus has two nucleoli ; the suspensor consisting of two cells, is in the act of forming a bipartition across its upper cells.

Fig. 25.—The embryo proper has four cells, the suspensor five. Va-cuoles are appearing in the cells of the latter.

Fig. 26.—The embryo proper consists of eight cells, and the suspensor of four. Traces of "antipodal" mass still evident.

Fig. 26a.—Transverse section of embryo at this stage.

Fig. 27.—The suspensor possesses three cells, of which the middle one has just divided, but the nuclei are not yet completely seprated.

Fig. 28.—A slightly older stage than the last. Each of the four sus-pensor cells has become doubled ; vacuoles are appearing, and the cell-walls are swollen.

PLATE III.

Fig. 29.—The suspensor cells are elongating, sap cavities becoming established within. The embryo proper still preserves twelve cells, but would soon consist of sixteen, as division appeared in its lower cells.

Fig. 30.—By intercalary growth the suspensor has pushed its apex through the micropyle ; one more division has occurred above, but the limit is approaching, and the exhausted cells are acquiring large sap cavities. A series of tangential divisions has marked out an epiderma layer to the embryo proper (optical sec. cleared with KHO).

Fig. 31.—Similar section of older embryo similarly treated ; a second series of tangential divisions has appeared more internally, an the "sus-pensor" is shrivelling up, the protoplasm of its cells disappearing.

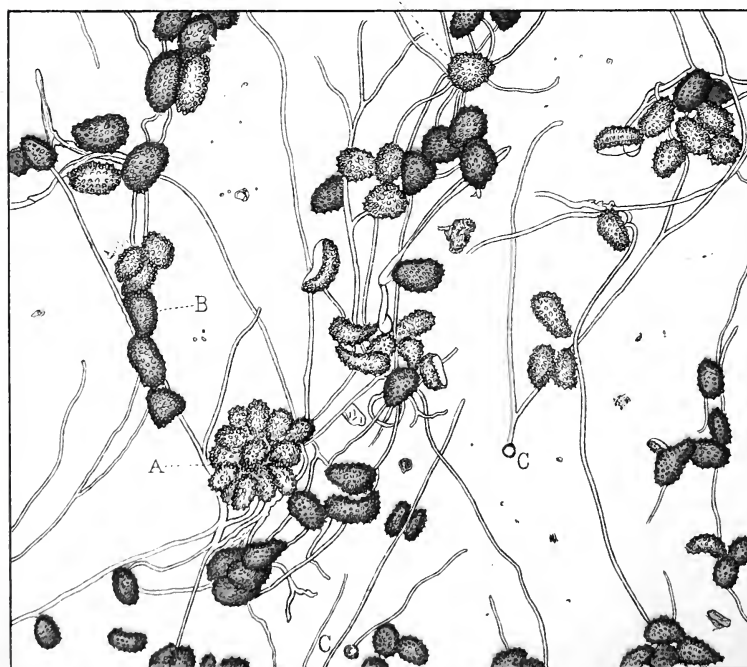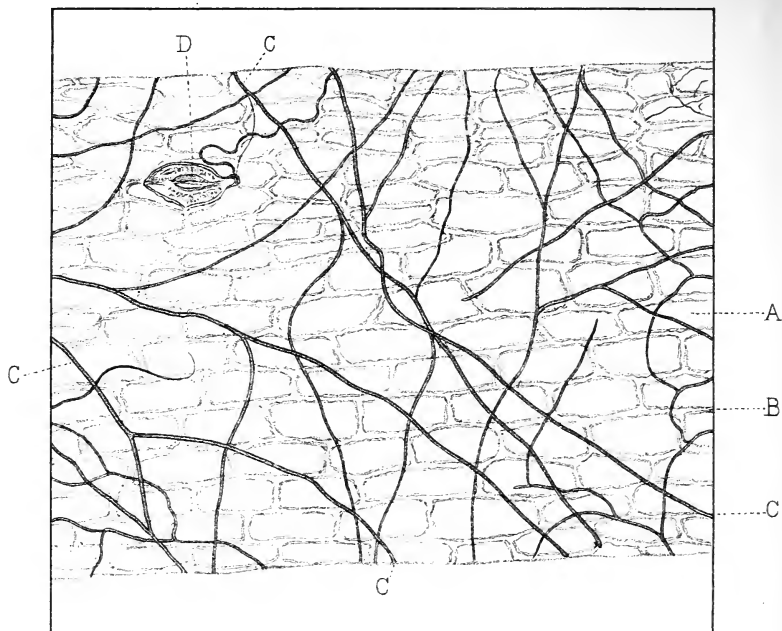Fig. 32.—External view of ripe seed torn from funiculus. Less magni-fied than rest.
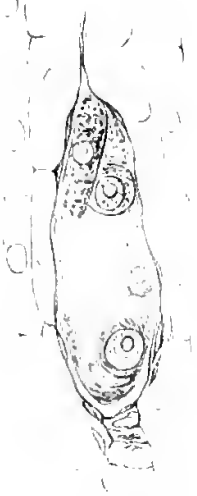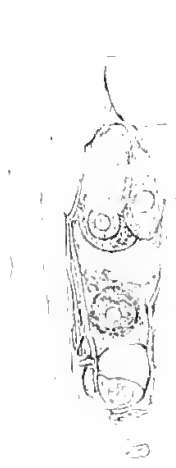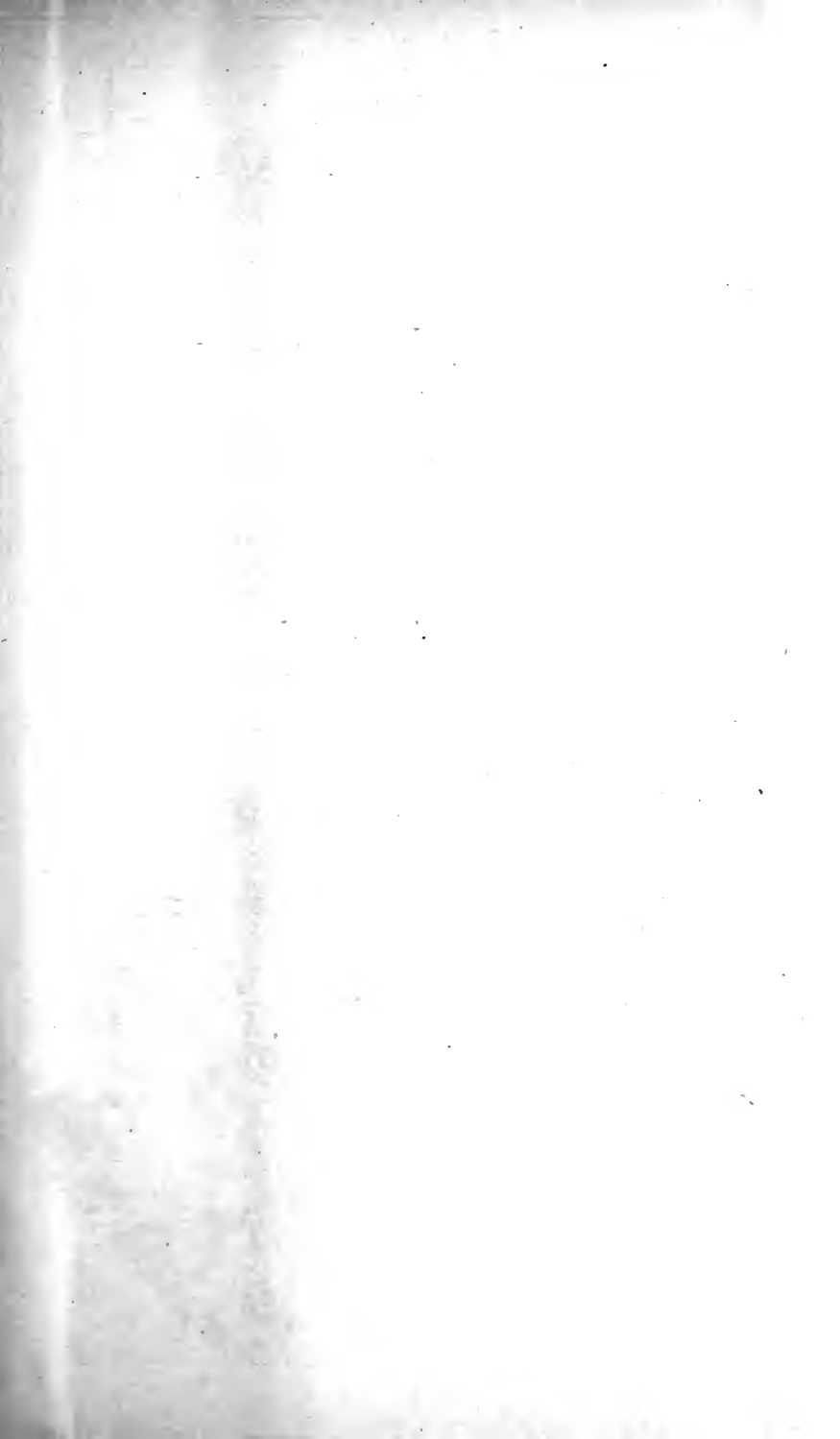
Fig. 15. Fig. 16. Fig. 17. Fig. 17 a. Fig. 18. Fig. 19. Fig. 20. Fig. 21. Fig. 22. Fig. 23. Fig. 24. Fig. 25. Fig. 26. Fig. 26 a. Fig. 27. Fig. 28.
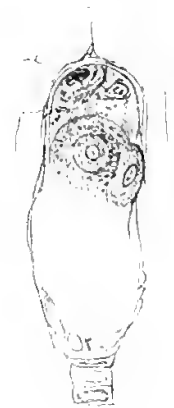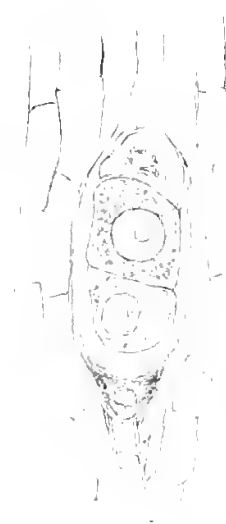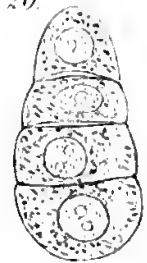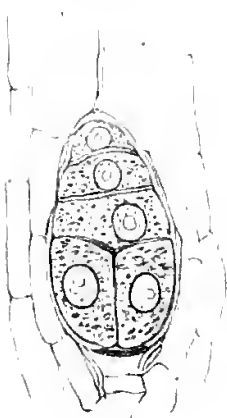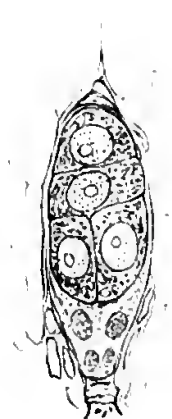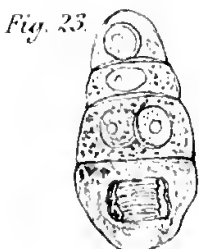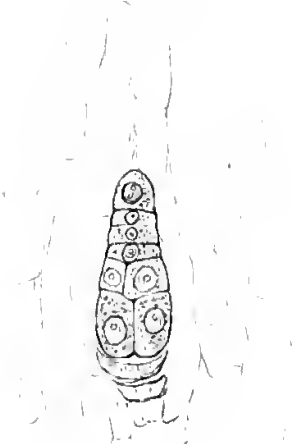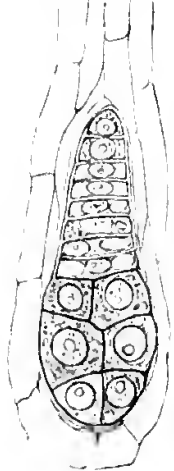
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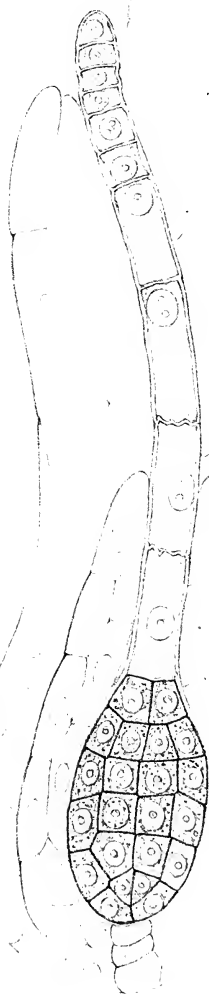
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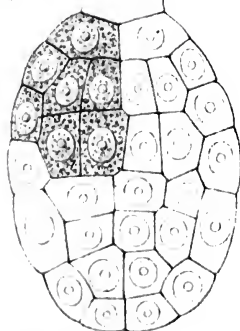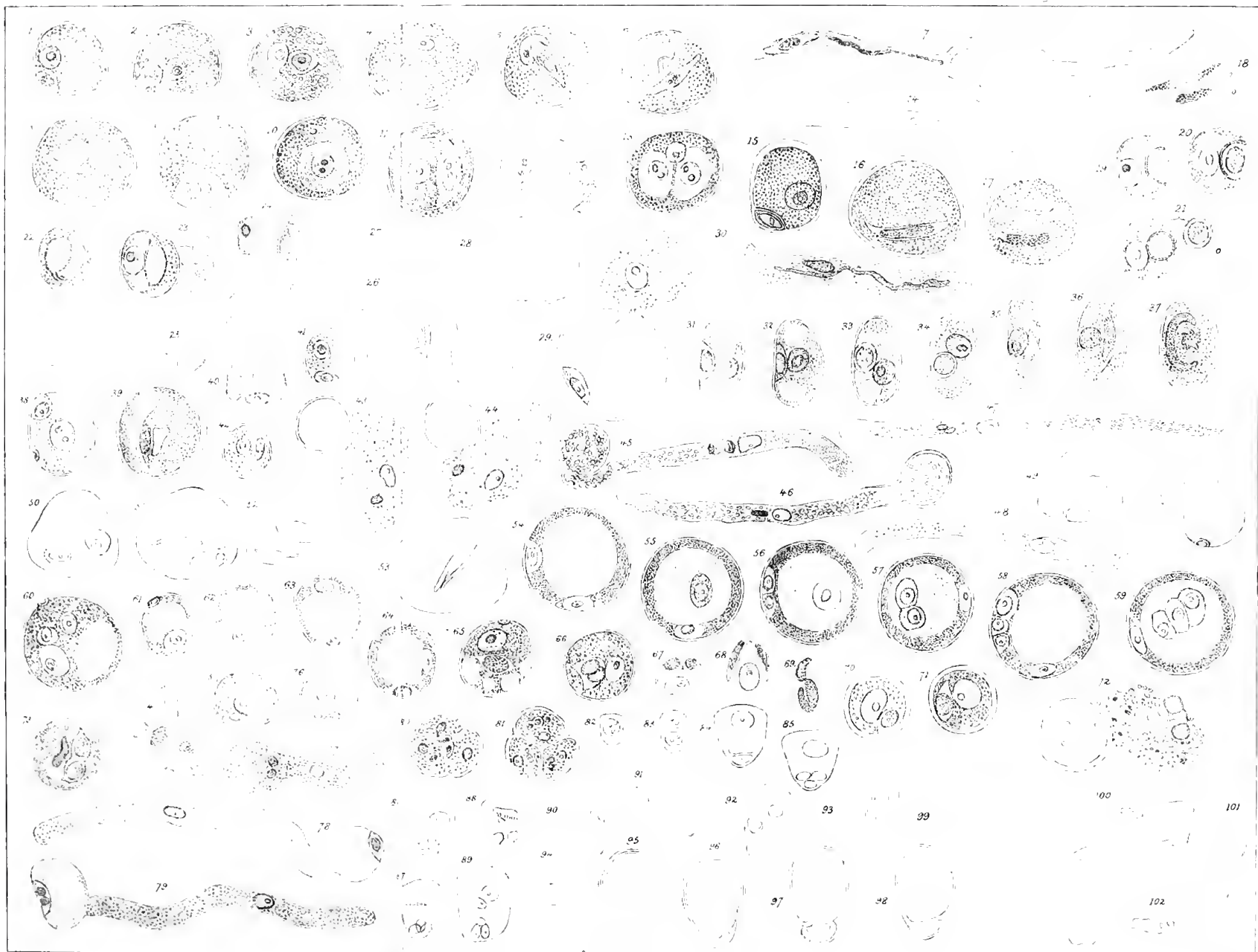
EXPLANATION OF PLATE IV,

Illustrating the notice of Elfving's researches on " Pollen-bodies of the Angiosperms."

Fig.   *Anthericum ramosum.*

1.—Young Pollen-body after division.   × 300.

2—4.—Development of the vegetative cell.   × 300.

5.—Metamorphosis of the nucleus of the large cell.   × 300.

6.—Ripe Pollen-body.   × 300.

7.—Branched tube.   × 230.

Anthericum liliago.

8.—Ripe Pollen-body, one nucleus star-shaped.   × 300.

Tulipa Gesneriana.   × 320.

9—11.—Development of the Pollen-body from formation of vegetative cell to being fully ripe.

12.—Vegetative cells pressed out.

13.—Duplicature of vegetative cell.

14.—Point of the Pollen-tube.

Ornithogalum pyramidale.   × 450.

15.—Vegetative cell with thick side walls.

16, 17.—Ripe Pollen-bodies.

18.—Pressed out vegetative cells; at *o* treated with osmic acid.

Leucojum æstivum.   × 400.

19.—Young Pollen-body after division.

20.—The same after treatment with osmic acid.

21.—The vegetative cell loosened ; at *o* with osmic acid.

22, 23.—Ripe Pollen-bodies ; *o*, a vegetative cell with osmic acid.

24.—Pressed out nucleus and vegetative cell; osmic acid.

25.—Pollen-tube.   Nuclei after one another.

Narcissus poëticus.

26.—Entering of the nuclei into the tube.

27.—The posterior nucleus divided.

Iris siberica.   × 350.

28, 29.—Crushed-out Pollen-bodies; in 28 the walls of the vegetative cell are seen, in 29 the whole cell.

Iris xiphium.   × 300.

30.—Tube end.   The nucleus of the larger cell is characteristically lengthened out.

Tradescantia virginica.   × 300.

31.—Young Pollen-body after division.

32—37.—Development of the bodies up to ripeness.

Convallaria multiflora.

38.—Formation of two vegetative cells.   × 350.

39.—Ripe Pollen-body.   × 400.

Asparagus officinalis.

40.—Arrangement of the vegetative cell.   × 600.

PLATE IV—*Continued.*

Fig.   *Sparganium ramosum.*   × 450.

41, 42.—Arrangement and first formation of the vegetative cell.

43, 44.—Crushed-out Pollen-bodies showing the metamorphoses of the vegetative nuclei.

45—48.—Pollen-tubes.   45. The vegetative nucleus in the act of dividing. 46. The vegetative nucleus has gone first, and has in 47 divided. 48. The vegetative nucleus has remained undisturbed.

Asphodelus albus.   × 350.

49—53.—Development of the Pollen-bodies from the first appearance of the vegetative cell until maturity.

Andropogon campanus.   × 400.

54.—Pollen-body after division.   55. Same seen at an angle of 90°.

56.—The vegetative cell divided.   57. Same seen from the side.

58.—Pollen-body with three vegetative cells.   59. The same seen at an angle of 90°.

60.—Both vegetative cells become free.

Bromus erectus.   × 350.

61, 62.—Pollen-bodies after division.

63—65.—Formation of the vegetative cell.

66.—Division of the vegetative cell.

67—69.—Metamorphosis of the vegetative cell and the larger nuclei up to the maturity of the Pollen-body.

Arum ternatum.   × 600.

70, 71.—First development of the Pollen-bodies after division.

72.—Pressed-out body; the division of the vegetative nuclei is only seen.

73.—Mature body; the spectacle-like united vegetatve cells are still maintained.

74.—Nuclei pressed out of mature bodies.

Butomus umbellatus.   × 300.

75.—First division of the Pollen-bodies.

76.—The vegetative nucleus has in addition divided.

77, 79.—Pollen-tubes; in 78 and 79 the partition walls are still maintained.

Juncus articulatus.   × 300.

80.—Pollen-body after division.   81. Older body.

Heliocharis palustris.   × 350.

82—94.—Development of the Pollen-body to maturity. 90 and 91 show the gradual resorption of the three nuclei situated in the narrow end.

95.—Normal thickening of the intine of a mature body.

96—98.—Peculiar thickenings of the membrane, which occasion the formation of false partition walls.

99—102.—Pollen-tubes.

Fig. 1.
Fig. 2.
Fig. 3.
Fig. 4.
Fig. 5.
Fig. 6.
Fig. 7.
Fig. 8'.
Fig. 8".
Fig. 8'".
Fig. 9.
Fig. 10.
Fig. 11.
Fig 12
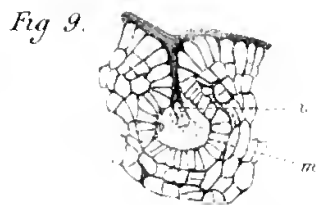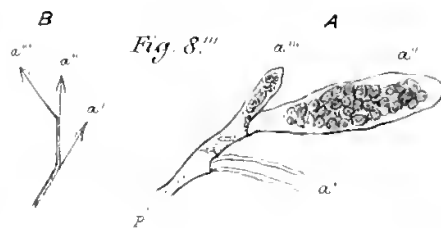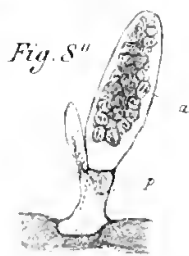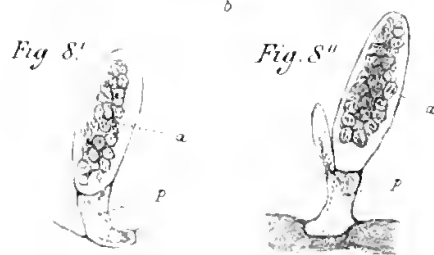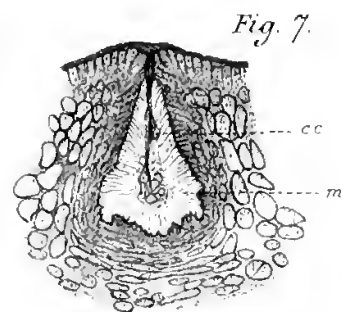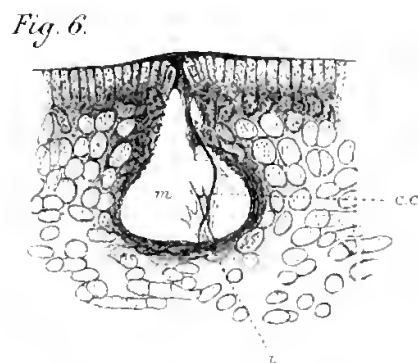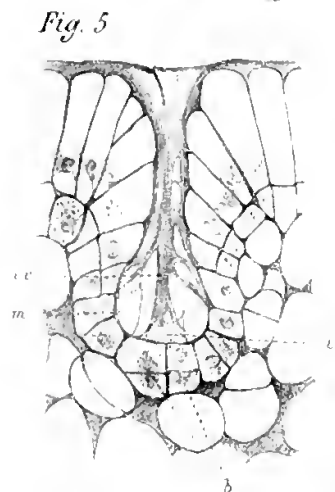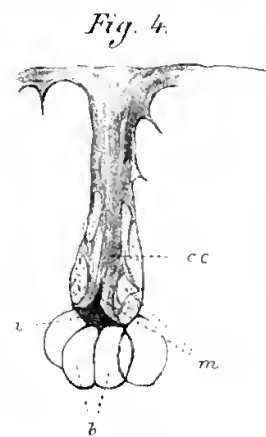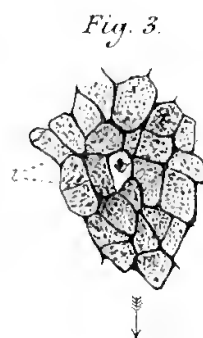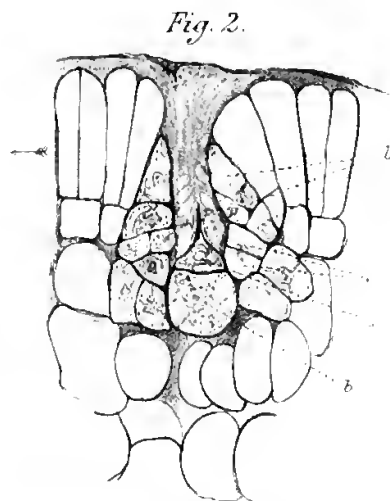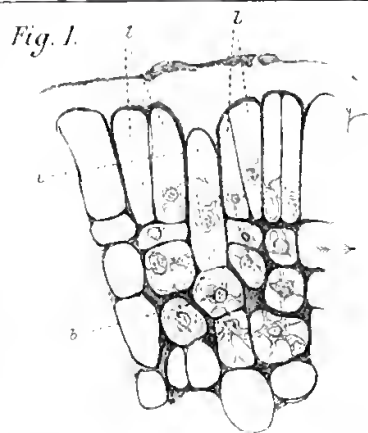
EXPLANATION OF PLATE V,

Illustrating Mr. F. O. Bower's Memoir on the Development of the Conceptacle in the Fucaceæ.

The following system of lettering has been used throughout—Initial cell = i. Basal cell = b. Limiting tissue = l. Cortical tissue = c. Central column = cc. Mucilage = m. Antheridial cell = a. Pedicel cell = p.

The first six figures are all from material treated first with dilute chromic acid. The remaining figures are from material preserved first in saturated solution of common salt (cf. text).

Fucus serratus.

FIG. 1 ($\frac{530}{1}$).—Part of a vertical longitudinal section passing through the lip which surrounds the depressed apex of a fertile branch. The initial cell has ceased to divide, and is surpassed by the surrounding tissue, a slight inclination is observable in the walls dividing the cells of the limiting tissue.

FIG. 2 ($\frac{530}{1}$).—Part of a similar section, with an older conceptacle. The initial cell has begun to shrink. The basal cell has not yet divided. The inclination of the walls of the limiting tissue is more pronounced than in fig. 1.

FIG. 3 ($\frac{530}{1}$).—A young conceptacle as seen from above, the protoplasm of the initial cell has shrunk, and its cell-walls swollen.

FIG. 4 ($\frac{530}{1}$).—Taken from a section similar to 1 and 2. This shows the initial cell more shrunk, the basal cell divided longitudinally. The alteration of the swollen cell-wall filling the cavity has begun.

FIG. 5 ($\frac{530}{1}$)—Part of a transverse section, with a conceptacle more advanced; it is dificult here to tell the limit between the part of the lining tissue, derived from the limiting, and that derived from the cortical tissue.

FIG. 6 ($\frac{120}{1}$).—Part of a transverse section, with an older conceptacle, the central column still attached. No formation of hairs as yet.

FIG. 7 ($\frac{120}{1}$).—Male conceptacle. Mucilage showing stratification and striation.

FIG. 8 i, ii, i ($\frac{530}{1}$).—Antheridia in various stages of development from a single cell of the lining tissue.

Halidrys siliquosa.

FIG. 9.—Vertical longitudinal section, with conceptacle.

Hymanthalia lorea.

FIG. 10.—Young conceptacle seen obliquely from above, showing initial hair protruding from the cavity.

FIG. 11.—Slightly older conceptacle in longitudinal section. Initial cell shrivelled. u = Cuticularised outer layer of swollen covering of the limiting tissue (cf. text, footnote).

FIG. 12 ($\frac{10}{1}$).—Branching antheridial hair, showing transition, on formation of antheridia, from monopodial to sympodial system of branching. The branches are numbered according to their ages, 1 being the oldest.

EXPLANATION OF PLATES VI & VII,

Illustrating Mr. J. E. Bloomfield's paper "On the Development of the Spermatozoa." Part I. ' Lumbricus.''

Fɪɢ.

1.—Testis of the earth-worm—a small example as seen under low power.

2.—Cells from the trabecular sustentaculum of the seminal reservoir.

3.—Portion of wall of a seminal reservoir with fusiform cells of the sustentaculum.

4.—Transverse section of the seminal reservoir, showing the penetrating blood-vessels.

5.—A portion of a seminal reservoir more highly magnified, showing the spermatospheres packed in the vascular sustentaculum.

6—10.—Cells (spermatospores) from a young testis eased : osmic acid and picrocarmine.

10ᴀ.—Drawn on a larger scale.

11—15.—Cells from young seminal reservoir elongating to form the sustentacular fibres.

16—20.—Spermatospores, dividing into two (young 'polyplasts ").

21, 22.—Into four.

23.—Three segments.

24.—Eight segments or spermatoblasts, drawn in fresh state.

25, 26.—Ditto, with acetic acid. 26 showing central protoplasm or blastophor (bl).

26ᴀ.—Sixteen spermatoblasts.

27, 28.—A polyplast. 27, with acetic acid. 28, fresh

29.—Similar polyplast on treatment with osmic acid and picrocarmine.

30, 31, 32.—Further stage in segmentation, fresh.

33, 34, 35.—Ditto, treated with osmic acid and picrocarmine.

36, 38.—To show the central blastophor (bl) at this stage, and in the following.

37.—Showing refractive cap on the spermatoblasts.

39.—Showing protrusion of fine filament.

40.—Same stage fresh.

41—46.—Progressive stages, consisting in an elongation of the nuclei of the spermatoblasts, h, n, t, head, neck, tail.

Fig. 3.

Fig. 2.
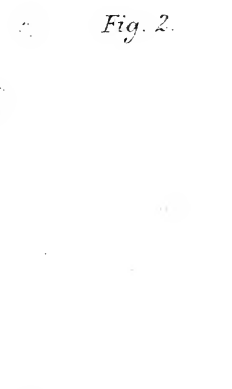
Fig. 5.
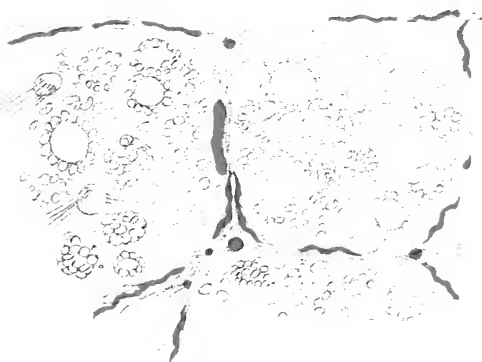
Fig. 1.

Fig. 4
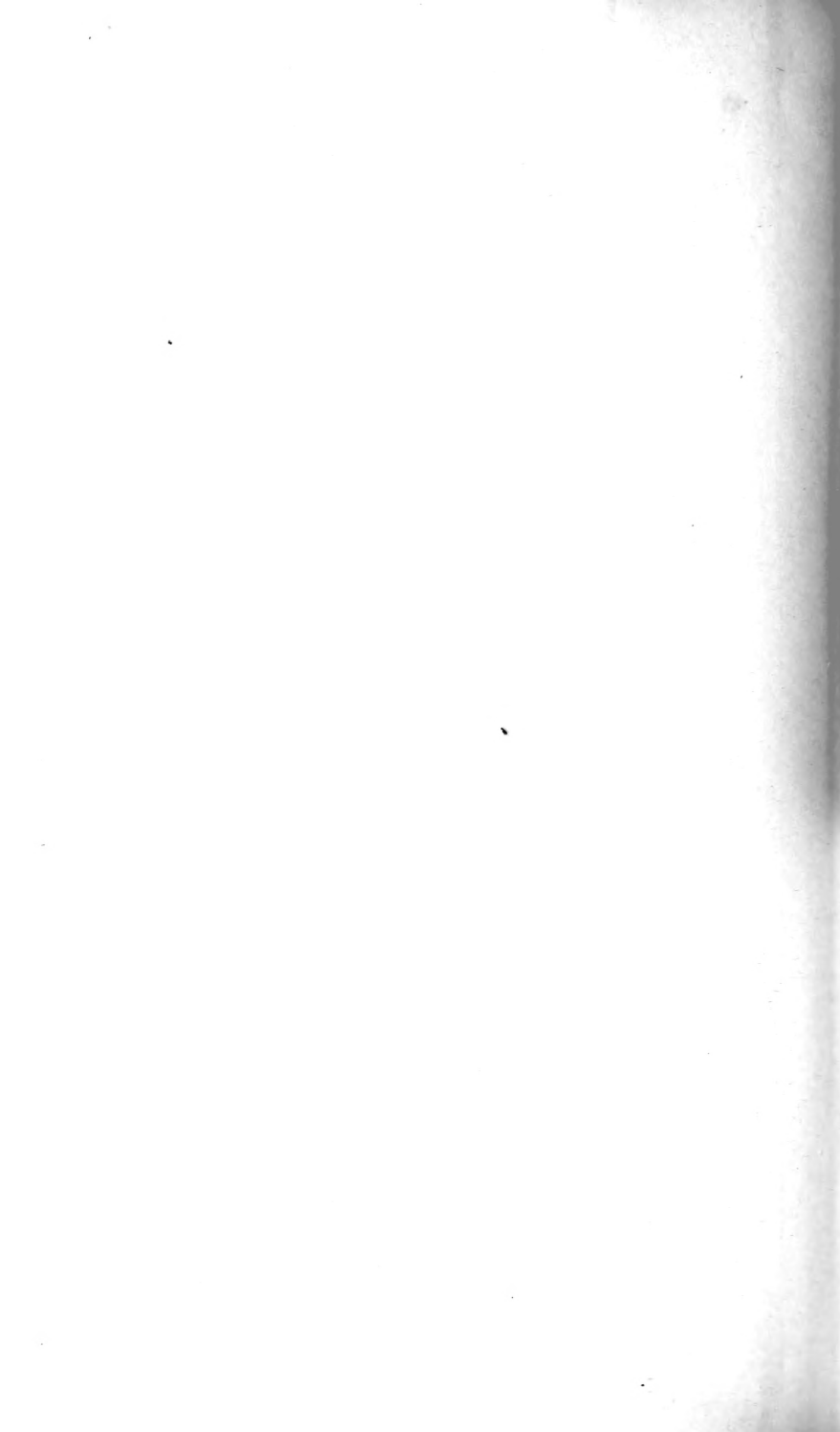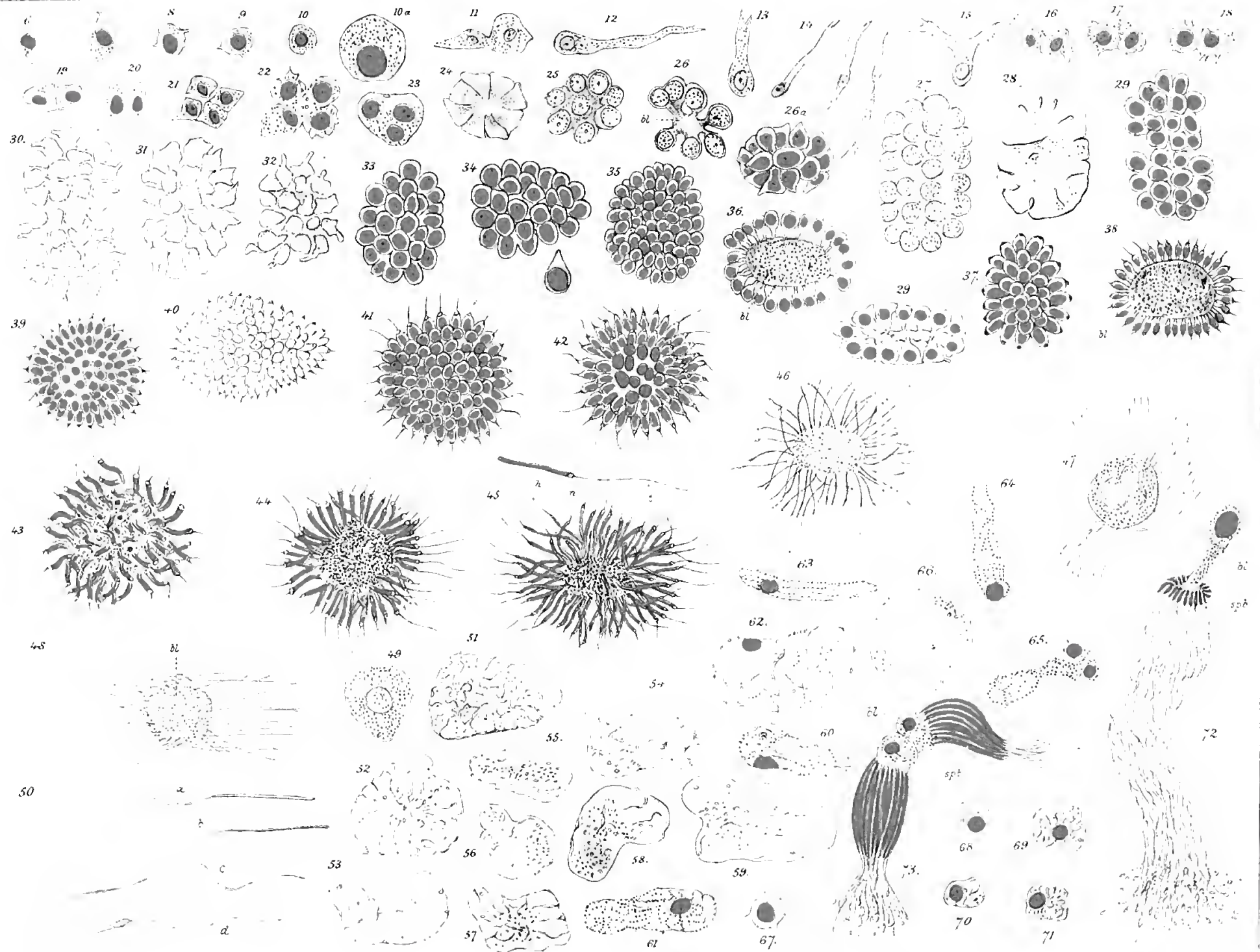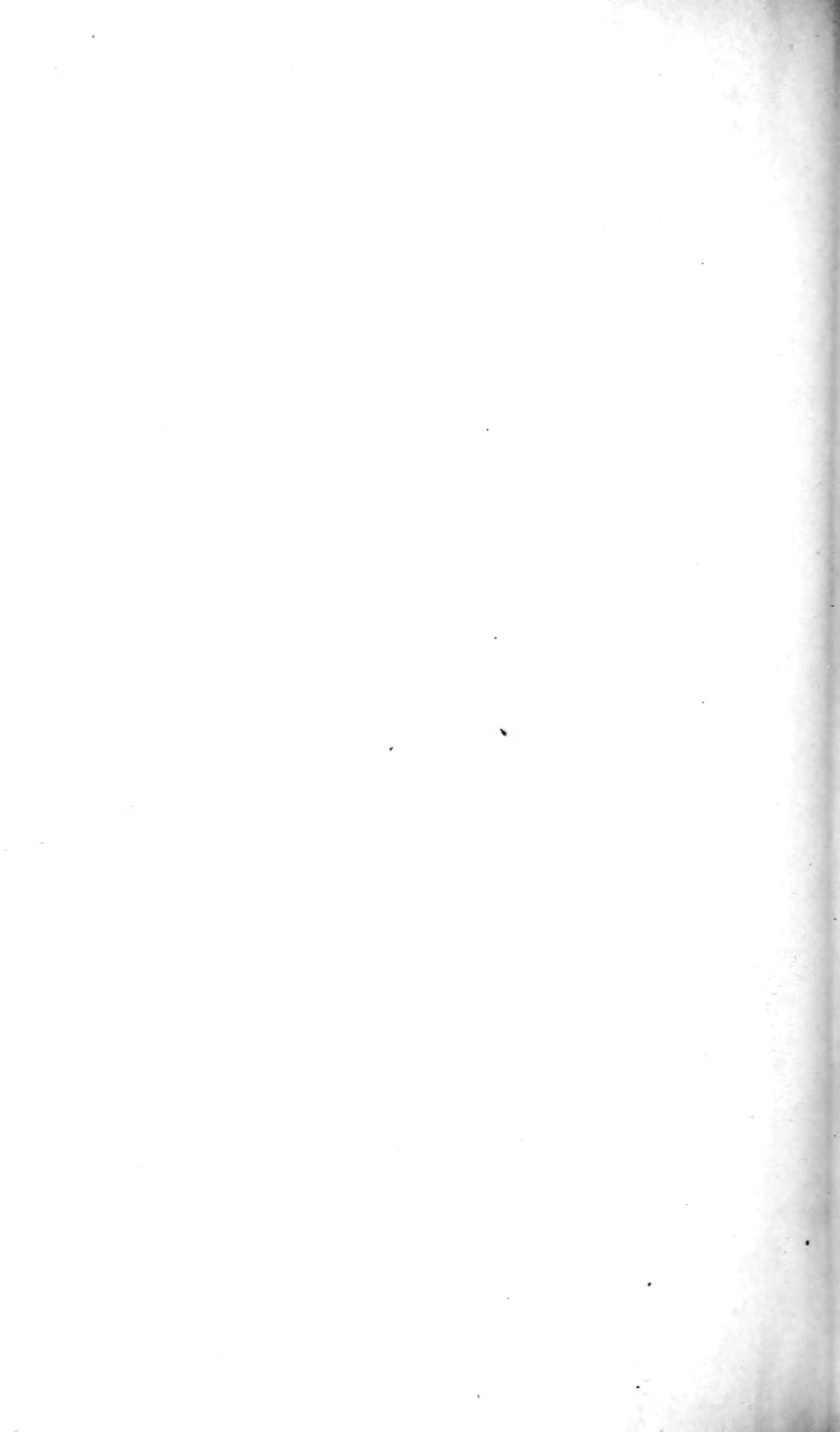
47, 48.—Spermatozoa mature, resting on central blastophor (*bl*).

49.—Isolated blastophor with vacuole.

50.—Spermatozoa seen under 10 immersion objective, *a, b, c,* mature ; *d,* from polyplast, fig. 46.

51—59.—Brown corpuscles of the seminal reservoirs exhibiting network, fresh.

60—65.—Ditto with osmic acid and picrocarmine.

66.—Similar brown corpuscle treated with acetic acid.

67—71.—Young brown corpuscle.

72.—Sperm polyplast of *Helix aspersa : bl.* blastophor, *spb.* spermatoblasts.

73.—Sperm polyplast of *Rena temporaria : bl.* blastophor, *spb.* spermatoblasts.

EXPLANATION OF PLATE VIII,

Illustrating Mr. Armaner Hansen's paper on the " Bacillus of Leprosy."

Figs. 1, 2, and 3.—Cells from tubercles with rod-shped bodies, fresh. Gundlach, No. 8.

Fig. 4.—Such cells treated with osmic acid, Gundlach No. 8.

Fig. 5.—A brown element with adhering articulated hreads after four days' cultivation.

Fig. 6.—From the border of one of the fungus growhs in the preparation of April 1st.

Fig. 7.—Brown elements coloured with methyl violel from a tubercle treated with osmic acid.

Fig. 8.—Bacilli coloured with methyl violet, from a secion of a tubercle hardened in absolute alcohol.

Fig. 1.

Fig. 3.

Fig. 2.

Fig. 4.

Fig. 5.

Fig. 6.

Fig. 7.
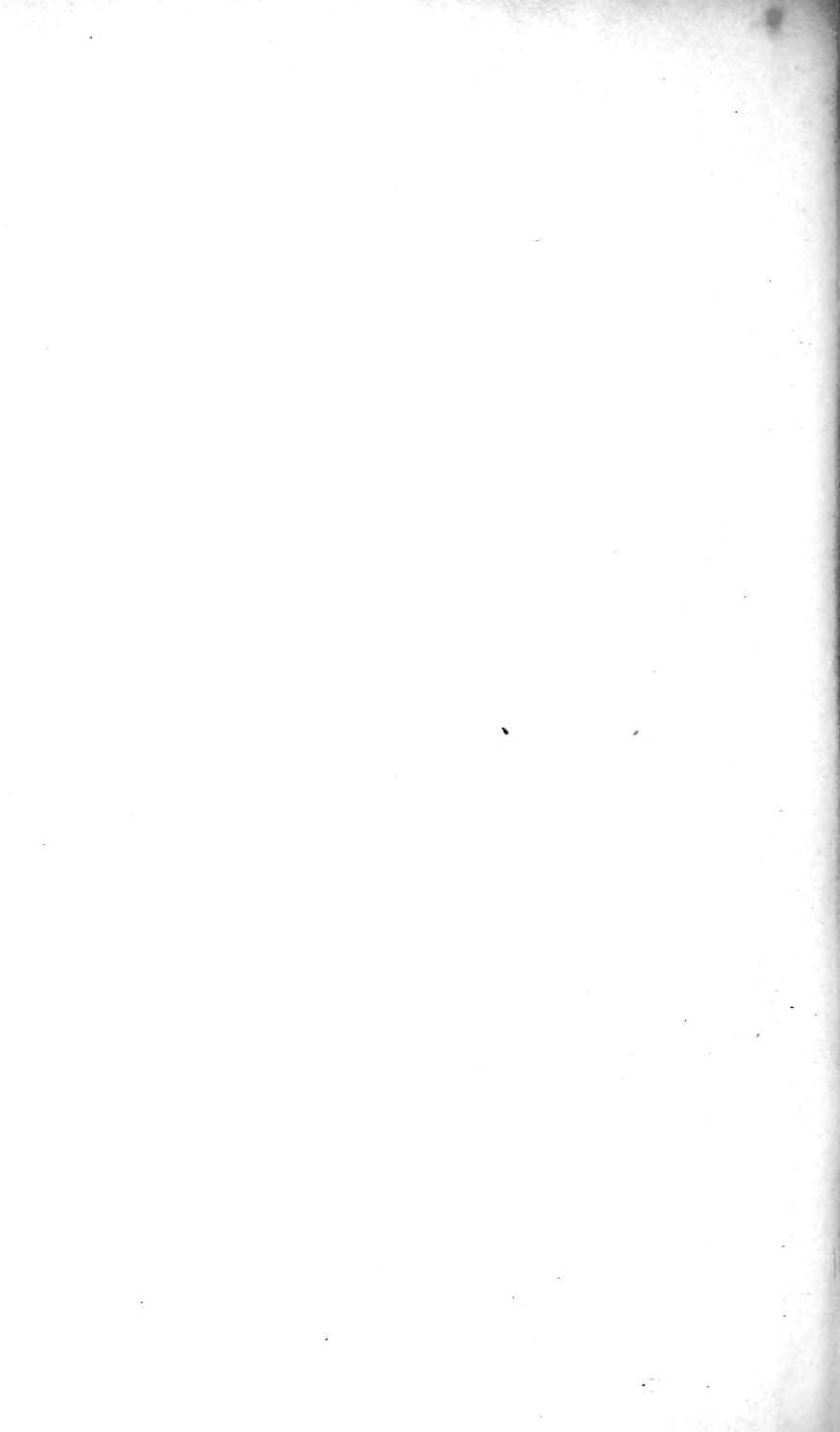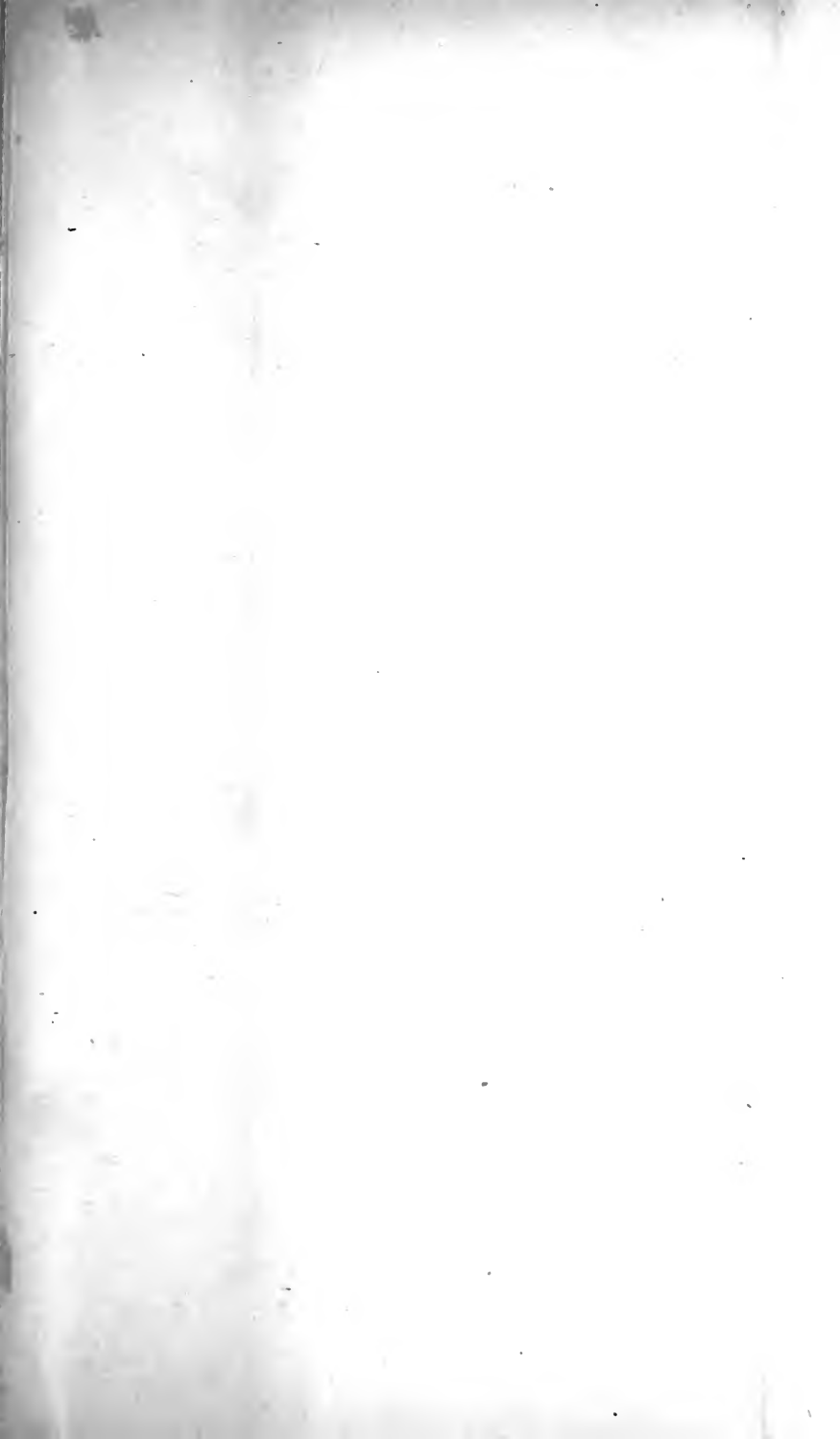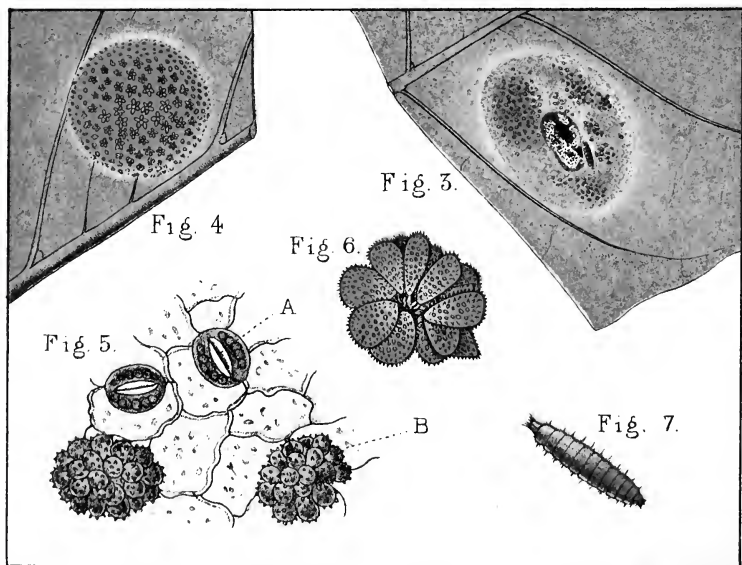
Fig 8.

Fig. 1.
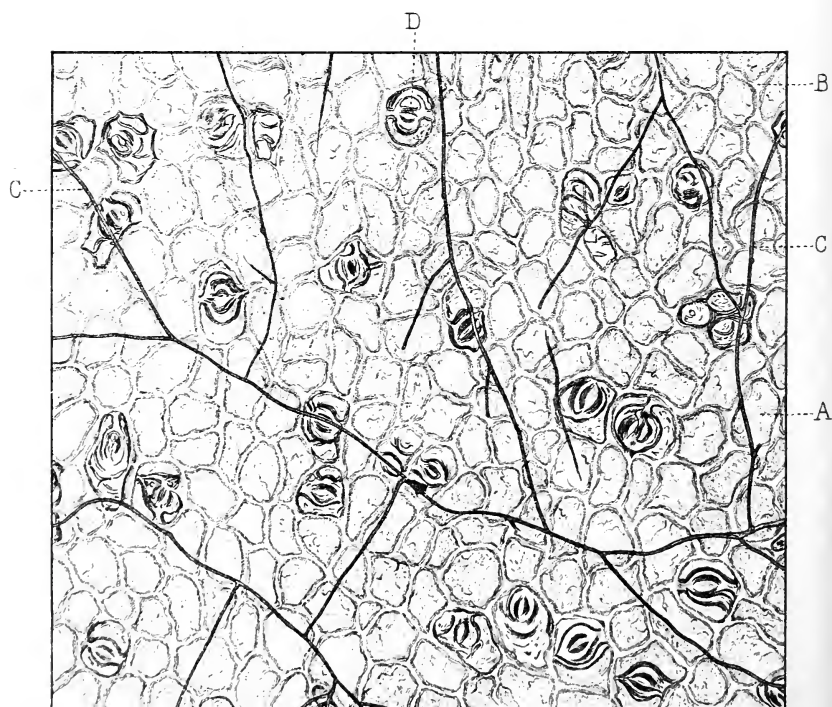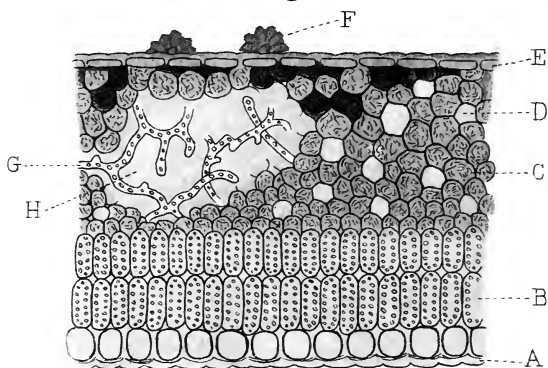
Fig 2.

Fig 4.

Fig. 3.

Fig 6.

Fig. 5.

Fig. 7.

HEMILEIA VASTATRIX.

Fig. 1.

Fig. 2.

HEMILEIA VASTATRIX.

Fig. 1.

Fig. 2.

HEMILEIA VASTATRIX.

Fig. 1.

Fig. 2.

Fig. 3

Fig. 1.

D

Fig. 2

D

F

E

Fig. 1.

Fig. 2.

D
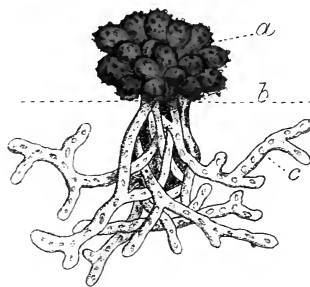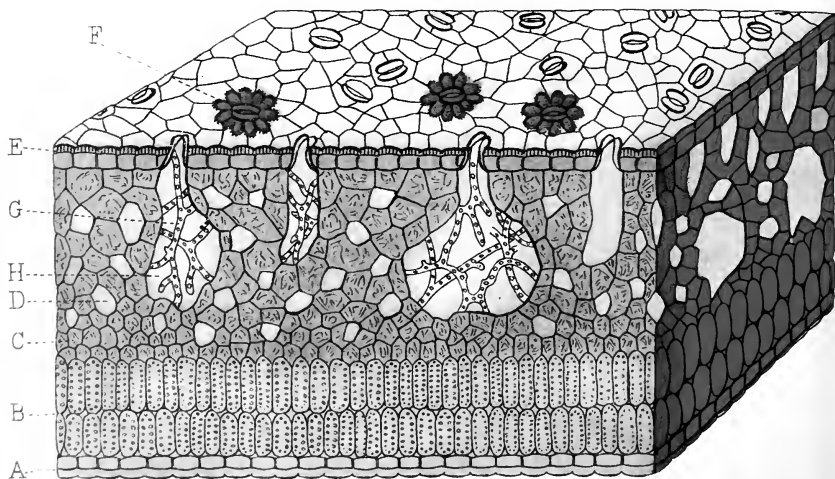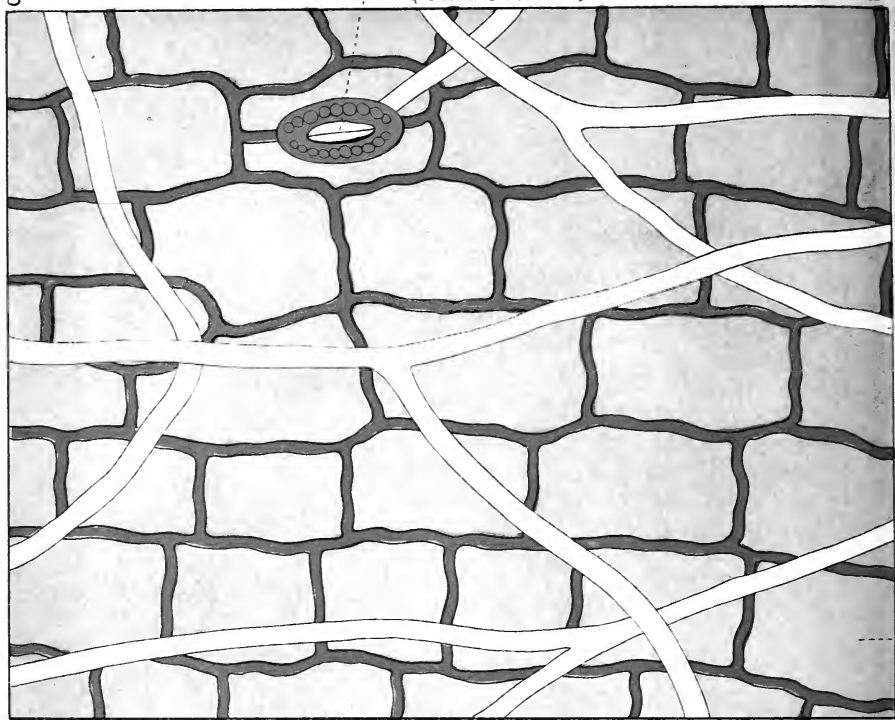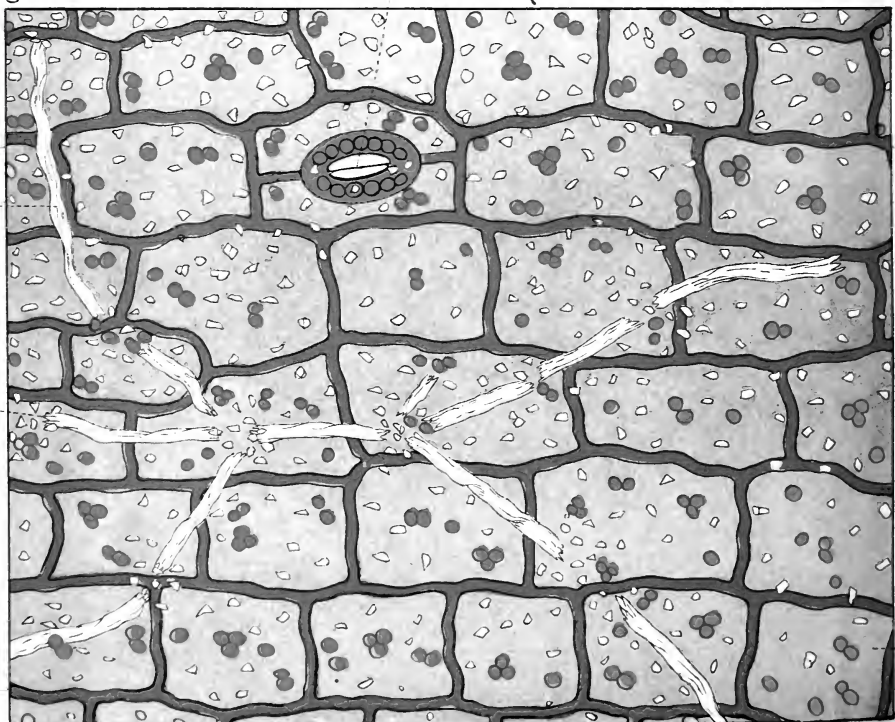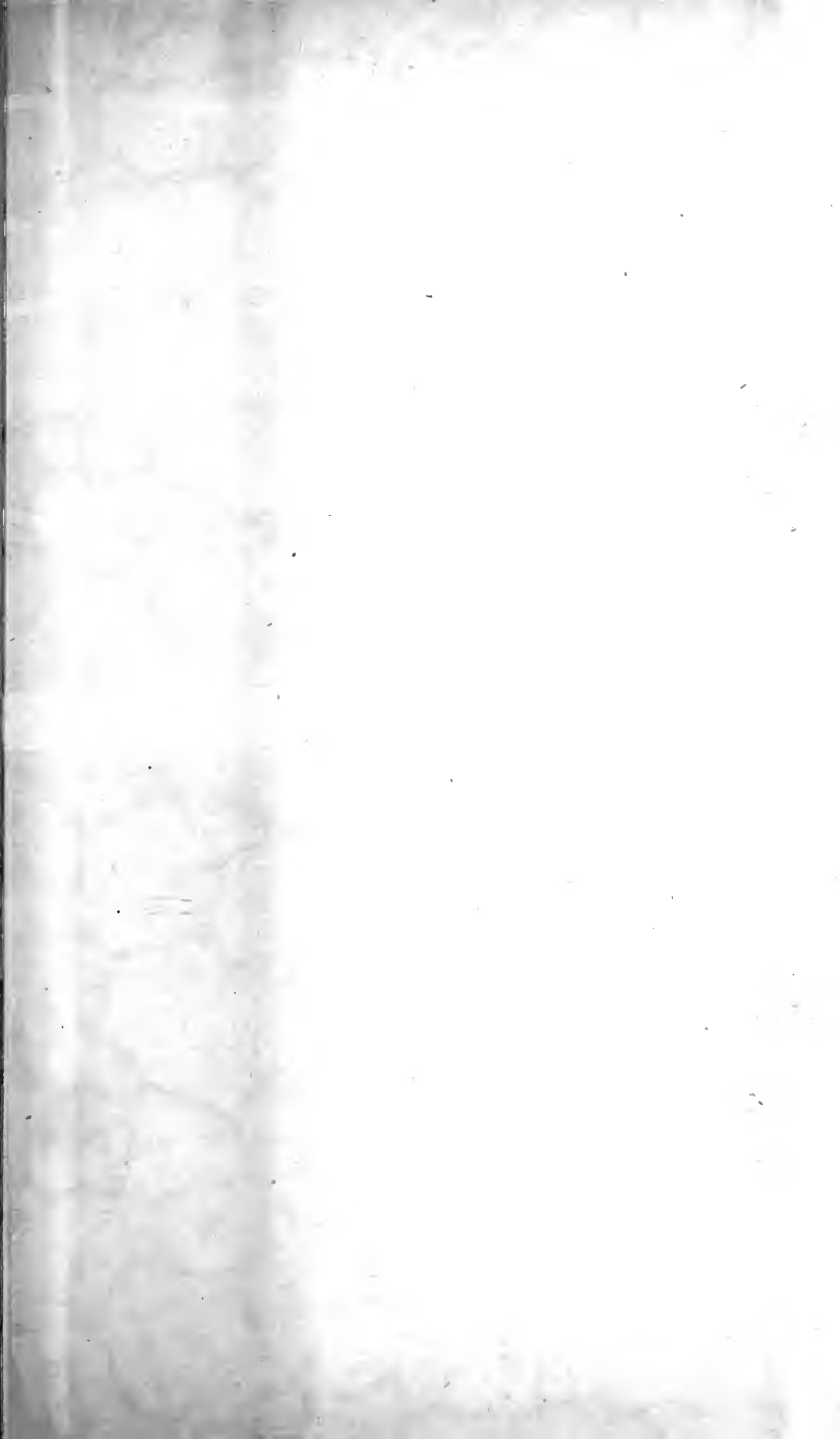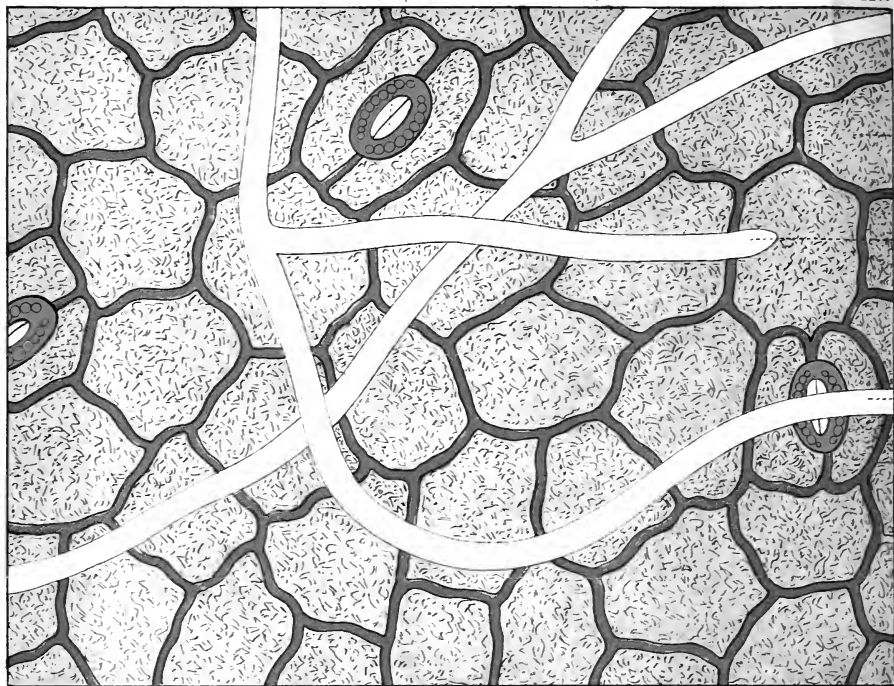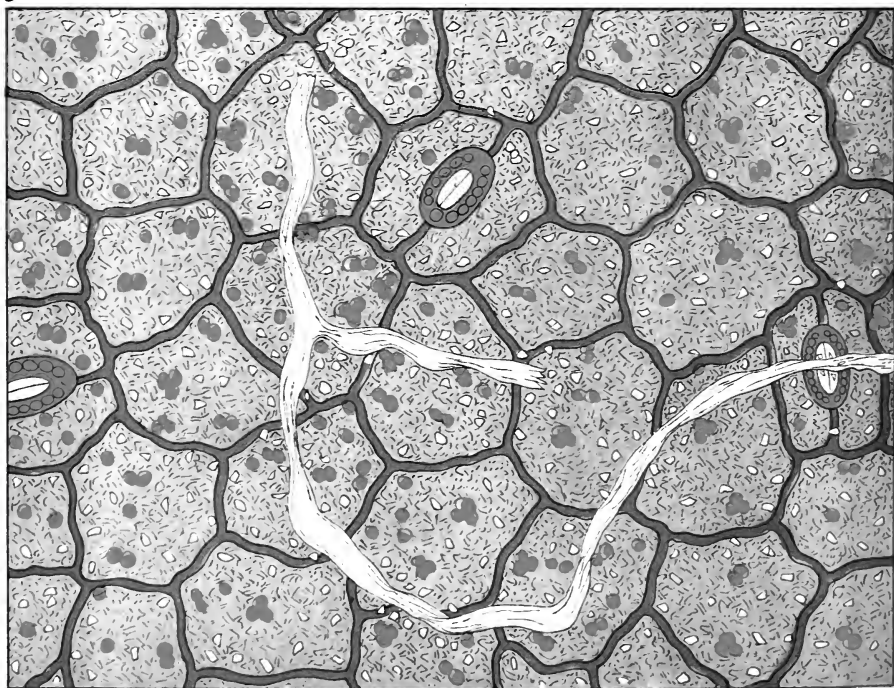
EXPLANATION OF PLATES IX—XIV,

Illustrating the paper on the "Coffee-leaf Disease of Ceylon," by W. T. Thiselton Dyer, M.A., F.L.S.

The whole of the drawings were made by D. Morris, M.A., F.G.S., for his forthcoming Handbook.

EXPLANATION OF PLATE IX.

Fig. 1.—Under side of coffee leaf, showing disease spots in various stages of development. The orange-coloured sporangia are arranged in numberless clusters on each spot. A. An old disease spot subsequently attacked by a second fungus (?*Aspergillus*). B. A disease spot traversed by nerves of the leaf. In such cases the filaments cross over the barriers formed by the nerves from one stoma to another, on the outside of the leaf.

Fig. 2.—Disease spots coalescing and forming an individual patch.

Fig. 3.—Portion of coffee leaf, twice natural size, with old disease spots in the centre. The immature sporangia are orange coloured, whilst the more mature are colourless. The larva of a male dipterous insect is represented feeding on the sporangia. See fig. 7.

Fig. 4.—Disease spot, magnified about ten times, showing the arrangement of the sporangia in clusters.

Fig. 5.—Portion of under surface of coffee leaf magnified, so as to show :—A. Stomata. B. Cluster of sporangia coming through and occupying the area of a stoma. × 200.

Fig. 6.—Single cluster of sporangia. The more mature have fallen off, exposing their points of attachment. × 500.

Fig. 7.—Enlarged drawing of larva of dipterous insect, found feeding on sporangia of *Hemileia*. See Fig. 3.

EXPLANATION OF PLATE X.

Fig. 1.—Sporangia found on fallen leaves and moist surfaces under coffee trees, giving rise to wide-spreading mycelial filaments. × 250.

Fig. 2.—Sporangia sown on glass slide, and kept in a moist atmosphere for thirty-six hours. Mycelial filaments abundantly produced. A. Unbroken cluster of ripe sporangia. B. Detached orange-coloured sporangia. c. Sporidia escaped from the sporangia, developing filaments. c. Sporidia developing filaments whilst still enclosed in the sporangia. × 600.

SHEPHEARDIA TÆNIFORMIS.
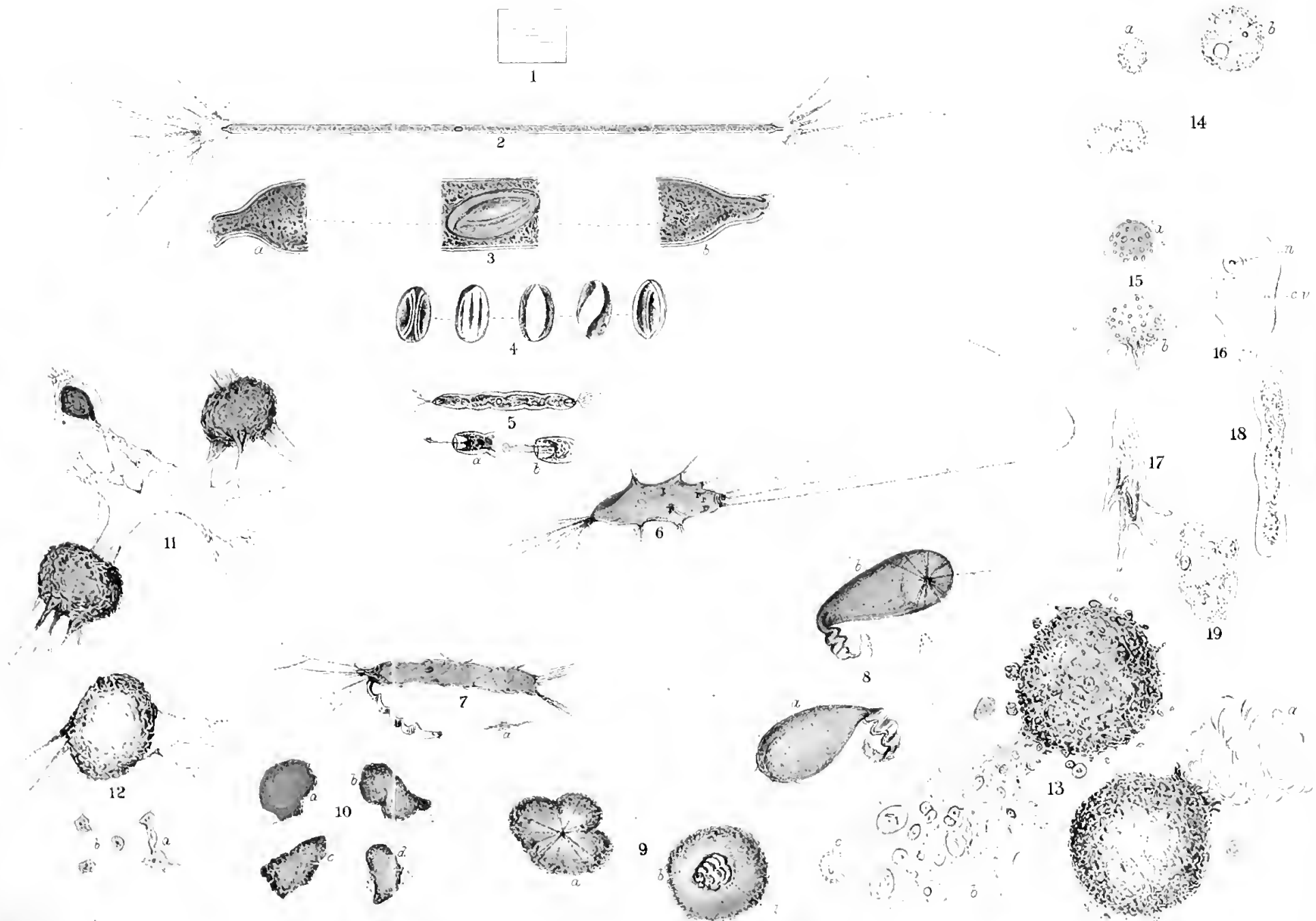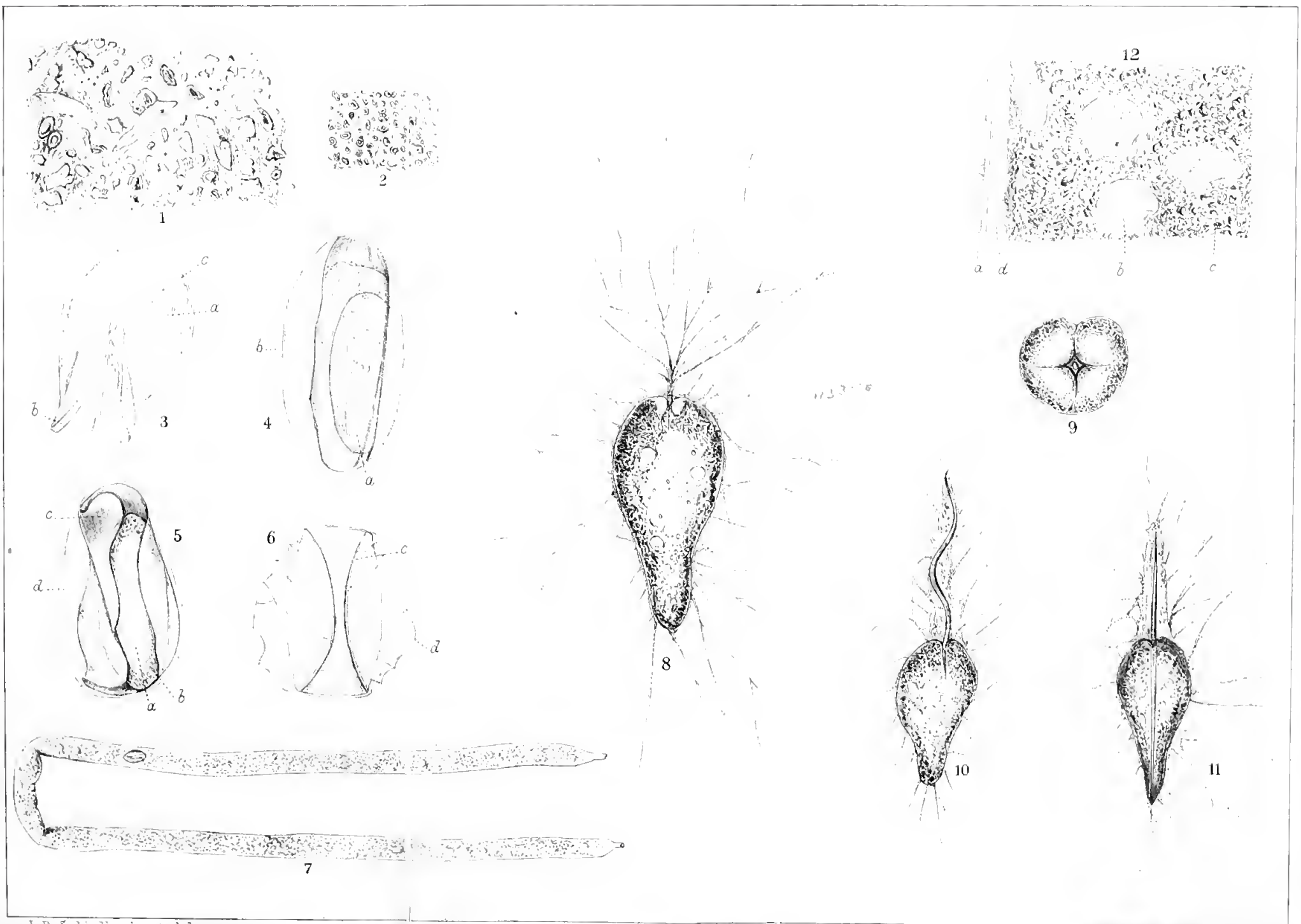
1–7 SHEPHEARDIA   8–12 LIEBERKUHNIA

EXPLANATION OF PLATES XV & XVI,

Illustrating Mr. J. D. Siddall's "Memoir on Shepheardella, an undescribed Type of Marine Rhizopoda; with a few Observations on Lieberkühnia."

PLATE XV,

Illustrating *Shepheardella tæniformis* life-history.

Fig. 1.—Three specimens drawn natural size.

Fig. 2.—A living *Shepheardella*, with pseudopodia extended from the two end apertures, and also from the investing layer of sarcode. Oval nucleus near the middle.   × 15 diameters.

Fig. 3 *a. b.*—The middle and two ends of same specimen, showing nucleus, apertures, integument, and yellowish granular sarcode. × 170 diams.

Fig. 4.—The varying appearances presented by the nucleus as it is carried along by the rotating protoplasm (*sarcode*).   × 120 diams.

Fig. 5.—A very small and somewhat abnormal specimen.   × 60 diams. *a.* and *b.* The end showing aperture and first protrusion of pseudopodia.

Fig. 6.—A many-apertured form, probably "*Shepheardella*," in first stage of "breaking up." Pseudopodia drawn to actual length.   × 27 diams.

Fig. 7.—Fig. 2 at nine a.m., December 18th. *a.* Detached portion of sarcode.

Fig. 8 *a. b.*—Fig. 2 at seven p.m., December 19th. Viewed from both sides of the cell.

Fig. 9 *a. b.*—Fig. 2 at nine a.m., December 20th. Viewed from both sides of the cell. Sarcode all *naked*, having been exuded from the wrinkled empty integument *i.*

Fig. 10 *a. b. c. d.*—The *naked* sarcode of another "*Shepheardella*," which, having passed through preliminary alterations in form, somewhat similar to those represented by Figs. 2, 7, 8, and 9, broke up into four separate portions on December 17th at nine a.m., twenty-four hours previously having been just as Fig. 9 in form, but still enclosed in its integument.

Fig. 11.—Fig. 10 at 10.30 p.m., December 17th.

Fig. 12.—Fig. 10 at nine a.m., December 18th. *a. b.* Detached amœboid portions. Figs. 7 to 12 × 20 diameters.

Fig. 13 *a. b. c.*—Fig. 10 on December 27th. Breaking up. *a.* The empty integument. *b.* A protruded mass of still sarcode, containing a number of nucleated granules. *c.* One of the latter which has developed Actinophrys-like rays.

Fig. 14 *a. b.*—Actinophrys-form some days after liberation.   × 250 diams. *a.* Dividing into three separate individuals. *b.* Large example, containing nucleus and contractile vesicle.

Fig. 15.—A minute portion of Fig. 12, detached on December 18th, containing a large number of granules exceedingly small, yet much larger and more definite in form than the ordinary granules of the sarcode.

a. As first given off. *b.* Its appearance on December 20th, showing "lobose" pseudopodia and granules given off from it. By January 25th it had discharged all the granules, which could not be distinguished from the other organic matter on the slide.

FIGS. 16, 17, 18, and 19.—Amœbæ, as given off from *Shepheardellæ;* 16 containing nucleus. *n.* Nucleus. *c. v.* Contractile vesicle. 17. Another form, occasionally nucleated. 18. Non-nucleated, very active form. 19. Resting condition, as now assumed, four to seven weeks after liberation.

PLATE XVI,

Illustrating *Shepheardella tæniformis* histology, and *Lieberkühnia.*

FIG. 1.—Camera tracing of the subcutaneous layer of the sarcode of "*Shepheardella,*" mounted in glycerine jelly (without any other treatment) and containing, imbedded in its clear, structureless protoplasm, nucleated granules of various sizes, and large clear masses of firmer protoplasm. × 600 diams.

FIG. 2.—Camera tracing of a similarly mounted specimen, after treatment with osmic acid, alcohol, and picro-carmine, showing every granule contained in the clear protoplasm to be of definite form and nucleated. × 600 diams.

FIG. 3.—The nucleus of a glycerine-mounted specimen (only), showing the nucleus, *a,* proper, enveloped in a membrane, *b c.* × 300 diams.

FIG. 4.—The nucleus of another specimen, after treatment with acetic acid and carmine, showing the nucleus, *a,* proper, filled with granular protoplasm (stained deeply), and an irregular edged denser mass near its centre (nucleolus), the nucleus being *embraced* by a delicate membrane, *b.* × 300 diams.

FIG. 5.—Another nucleus, treated and mounted as Fig. 4, showing *a,* nucleus; *b,* the protoplasmic contents contracted upon its walls; *c,* the embracing membrane; and *d,* the very transparent enveloping sac. × 300 diams.

FIG. 6.—Another, mounted in glycerine jelly only, showing *c,* the embracing membrane; and *d,* the enveloping sac wrinkled upon it. × 300 diams.

FIG. 7.—*Shepheardella tænia,* from a living specimen. × 40 diams.

FIG. 8.—*Lieberkühnia Wageneri.* × 55 diameters.

FIG. 9.—Aperture of ditto. Base of the principal stem of pseudopodia, shown as an oval spot within the square mouth. × 55 diams.

FIG. 10.—The same specimen shortly after being transferred to the cell, showing the living worm entangled among the pseudopodia. × 38 diams.

FIG. 11.—The same, with the worm swallowed as far as possible.

FIG. 12.—Marginal portion of the same when mounted in glycerine jelly, viewed in optical section, showing transparent integument, *a*; vesicular nuclei, *b*; granular protoplasm, *c*; and subcutaneous layer of finer protoplasm, *d.* × 1000 diams.
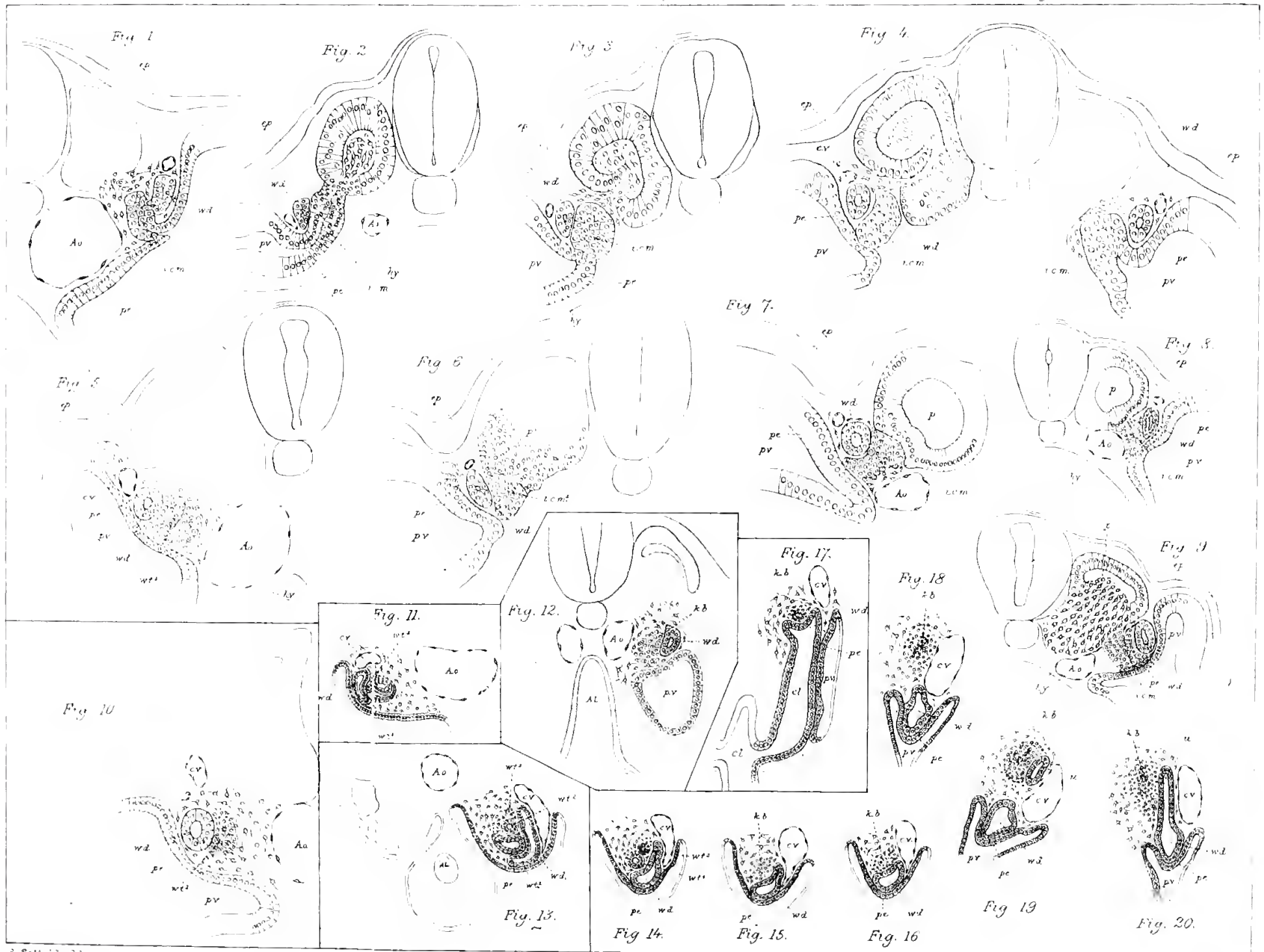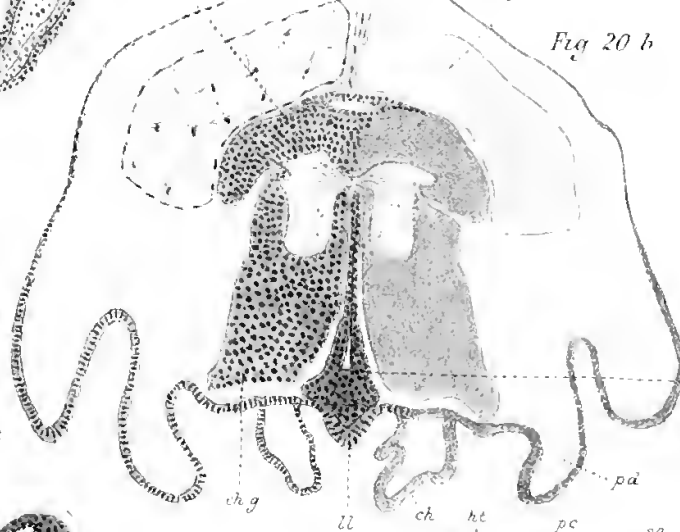
Fig. 21.
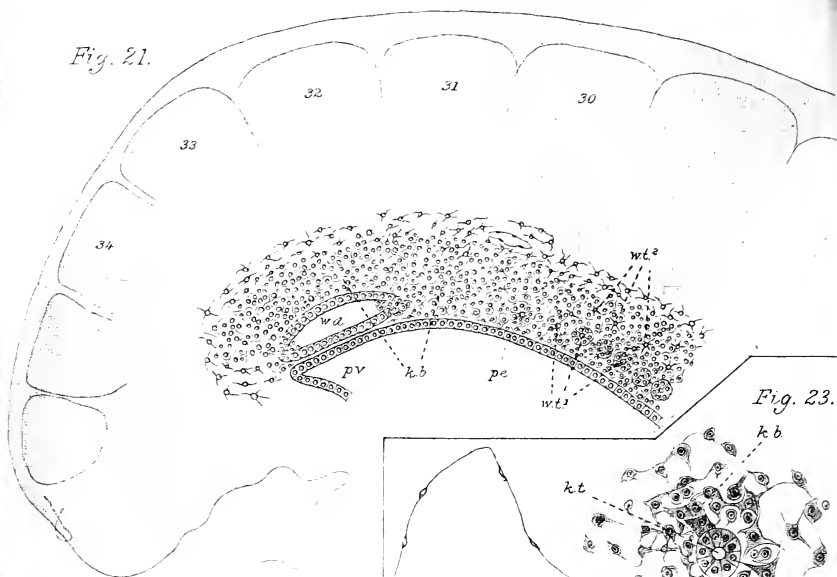
Fig. 22.

Fig. 23.
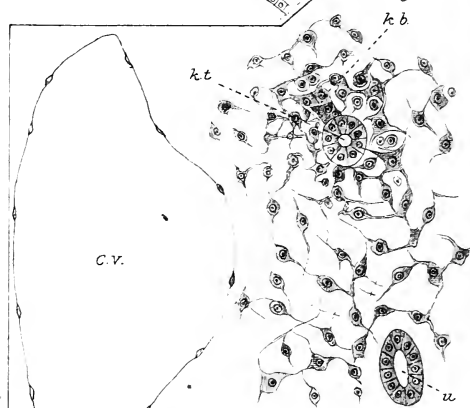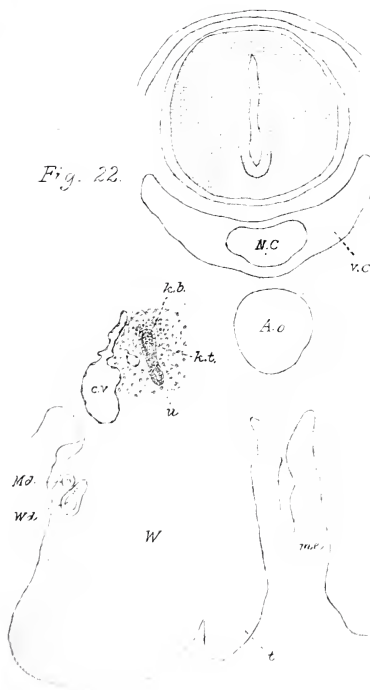
Fig. 24.
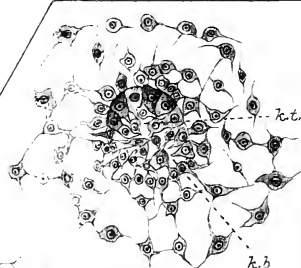
Fig. 25.
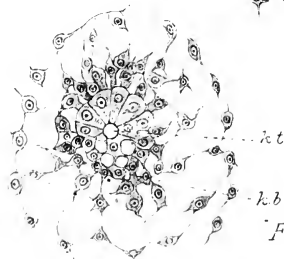
EXPLANATION OF PLATES XVII AND XVIII,

Illustrating Mr. Sedgwick's Memoir on " Development of the Kidney in its Relation to the Wolffian Body in the Chick."

Complete List of Reference Letters.

Ao. Aorta. *Al.* Alimentary canal. *cl.* Cloaca. *c. v.* Cardinal vein. *ep.* Epiblast. *hy.* Hypoblast. *i. c. m.* Intermediate cell mass. *i. c. m.'* Cell mass, which later becomes the intermediate cell mass. *m. e.* Mesentery. *M. d.* Müllerian duct. *k. b.* Kidney blastema. *k. t.* Kidney tubule. *n. c.* Notochord, *p.* Protovertebra. *p'.* Cell mass, which later becomes a protovertebra. *p. v.* Body-cavity. *p. e.* Peritoneal epithelium. *t.* Testis. *ve.* Vertebral body. *w.* Wolffian body. *w. b.* Wolffian blastema. *w. d.* Wolffian duct. *w. t.^{1}* Primary Wolffian tubule. *w. t.^{2}* Secondary ditto. *w. t.^{3}* Tertiary ditto.

Fɪɢ. 1.—Section between the fifteenth and sixteenth protovertebræ of a chick with twenty-three protovertebræ, showing the rudimentary continuation of the body-cavity into the intermediate cell mass and the connection which the latter has obtained with the Wolffian duct. The intermediate cell mass in anterior and posterior neighbouring sections has separated from the peritoneal epithelium.

Fɪɢs. 2, 3, 4, and 5.—Sections taken from a duck embryo with about thirty-two protovertebræ, illustrating the development of the Wolffian tubules. Hart. cam., ob. 4.

Fɪɢ. 2.—Section through the thirtieth segment, intermediate cell mass continuous with peritoneal epithelium, and containing a rudimentary prolongation of the body-cavity. Lumen of Wolffian duct doubtful.

Fɪɢ. 3.—Section through the twenty-ninth segment, intermediate cell mass separate from peritoneal epithelium.

Fɪɢ. 4.—Section through the twenty-sixth protovertebra, showing features similar to above.

Fɪɢ. 5.—Section through the twenty-second protovertebra ; commencing differentiation of Wolffian tubule.

Fɪɢs. 6—10.—Sections illustrating the more modified development of the Wolffian blastema, as seen in the chick behind the twentieth segment.

Fɪɢ. 6.—Section through a chick with twenty-six protovertebræ behind the four last-formed segment, showing the thick peritoneal epithelium, the Wolffian blastema in connection with the mass of cells which will become a protovertebra. Hart. cam., ob. 4.

Fɪɢ. 7.—Section through the twenty-ninth protovertebra of a chick with twenty-nine protovertebræ, showing the thick peritoneal epithelium and the Wolffian blastema in connection with the provertebræ. Hart. cam., ob. 4.

Fɪɢ. 8.—Section through the twenty-fourth segment of a chick with twenty-six protovertebræ, showing the Wolffian blastema separate from protovertebræ and thick peritoneal epithelium. Hart. cam., ob. 3.

Fig. 9.—Section through the twenty-fourth segment of a chick with twenty-nine protovertebræ, showing Wolffian blastema and thin peritoneal epithelium. Hart. cam., ob. 3.

Fig. 10.—Section through the twenty-ninth segment of a chick with thirty-four protovertebræ, showing the commencing development of a primary Wolffian tubule from Wolffian blastema. Hart. cam., ob. 4.

Fig. 11.—Section through a chick, end of third day or beginning of fourth, showing earliest appearance of a secondary tubule. Hart. cam., ob. 3.

Fig. 12.—Section through the thirty-second protovertebra of a chick with thirty-four protovertebræ, showing the kidney blastema.

Figs. 13—17.—A series of sections from the hind end of a chick of fourth day, illustrating the continuity of the Wolffian body with the cells forming the kidney blastema. Hart. cam., ob. 3.

Fig. 13.—Last section, in which a tertiary tubule was seen.

Fig. 14.—Last section, in which a secondary tubule was seen. The tubules in figs. 13 and 14 are contiguous.

Fig. 15.—Next section but one behind fig. 14.

Fig. 16.—Next section but one to fig. 15.

Fig. 17 a.—Section some distance behind that drawn in fig. 16.

Figs. 15, 16, and 17 show kidney blastema.

Fig. 17 shows opening of Wolffian duct into horn of cloaca.

Figs. 18—20.—Sections through a slightly older embryo than that from which above series was taken. Hart. cam., ob. 3. Showing (fig. 20), ureter opening into Wolffian duct, with shifted kidney blastema lying just internal to it.

Fig. 19.—Showing developing ureter and kidney blastema.

Fig. 20.—Section just anterior to ureter through the anterior end of the kidney blastema.

Fig. 21.—A longitudinal vertical section through the hind end of a four-day chick, showing continuity of kidney blastema with hindermost part of Wolffian blastema, in which the development of Wolffian tubule is taking place. No line of demarcation can be drawn between the two.

Fig. 22.—Section through a chick of seventh day or late in sixth, showing the portion of the ureter (*u.*) and its dorsal dilatation (*v. t.*) with regard to the Wolffian body (*w.*).

Figs. 23 and 24 are from sections of the chick from which fig. 22 was taken.

Fig. 23.—Section next but one to fig. 22. It shows the kidney tubule dorsal to the ureter, surrounded by the blastema.

Fig. 24.—Section next to fig. 22, shows the dilated termination of the kidney tubule and the continuity of its lining cells with those of the kidney blastema.

Fig. 25.—From a section through the kidney of an eight-day chick, showing the termination of a kidney tubule. It presents the same feature as fig. 24.
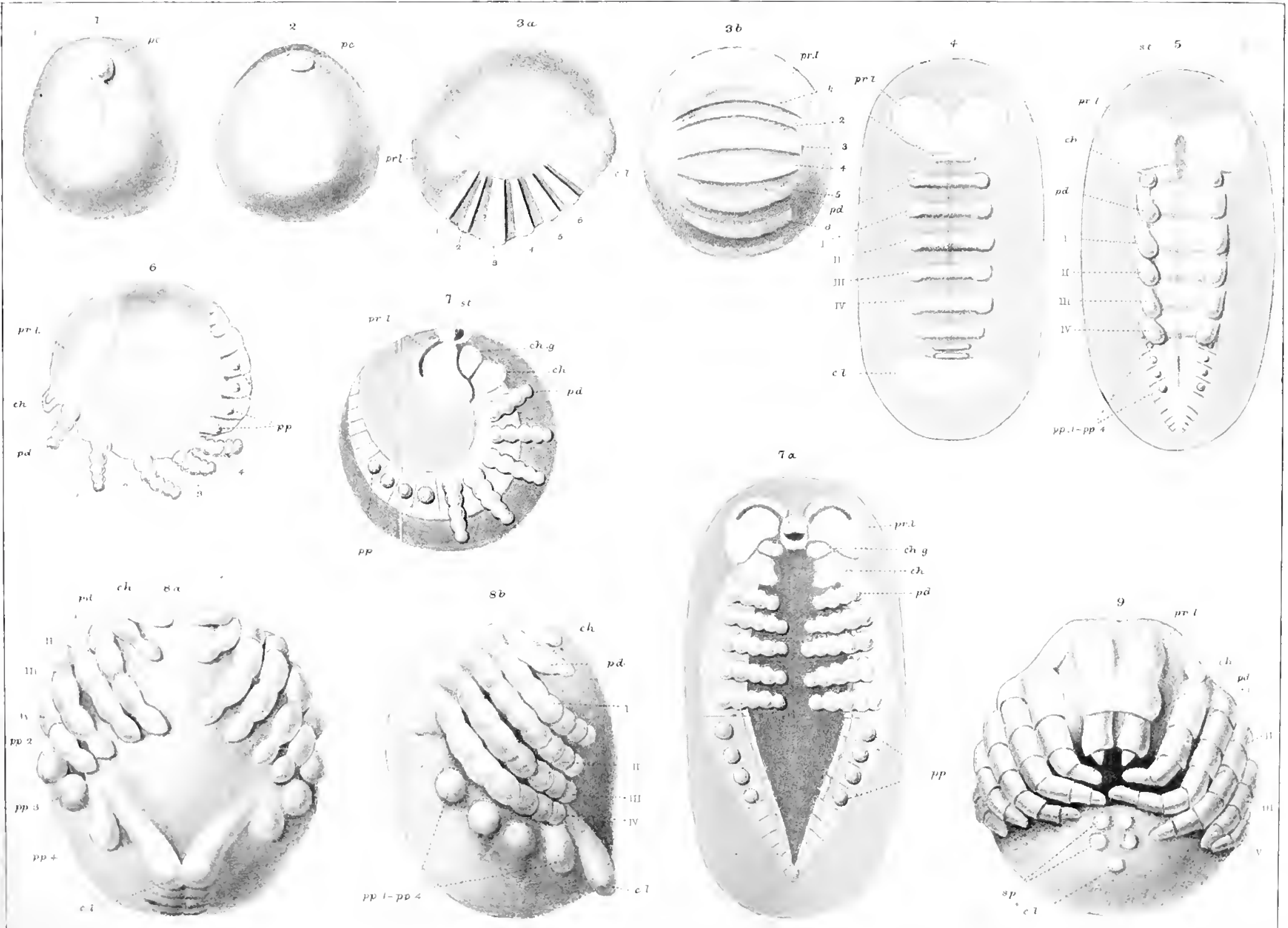
Fig. 10.
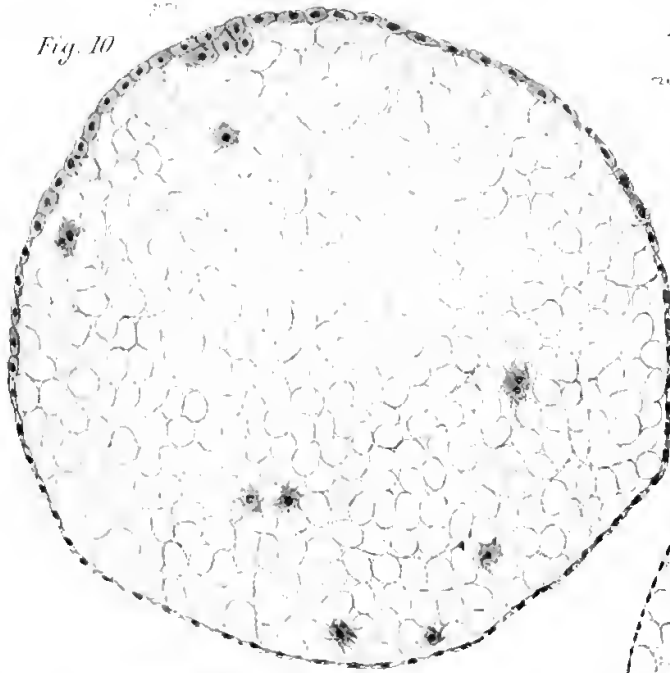
Fig. 11.
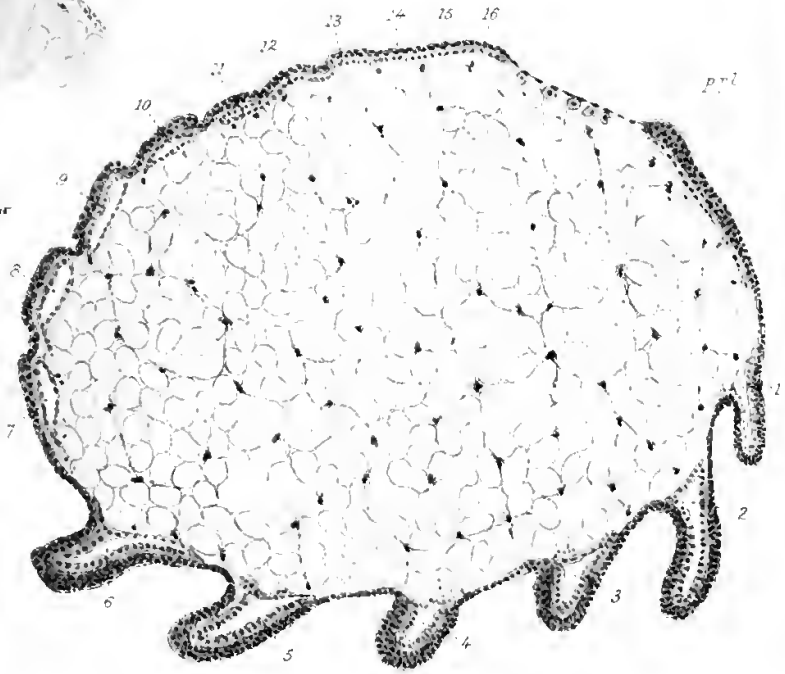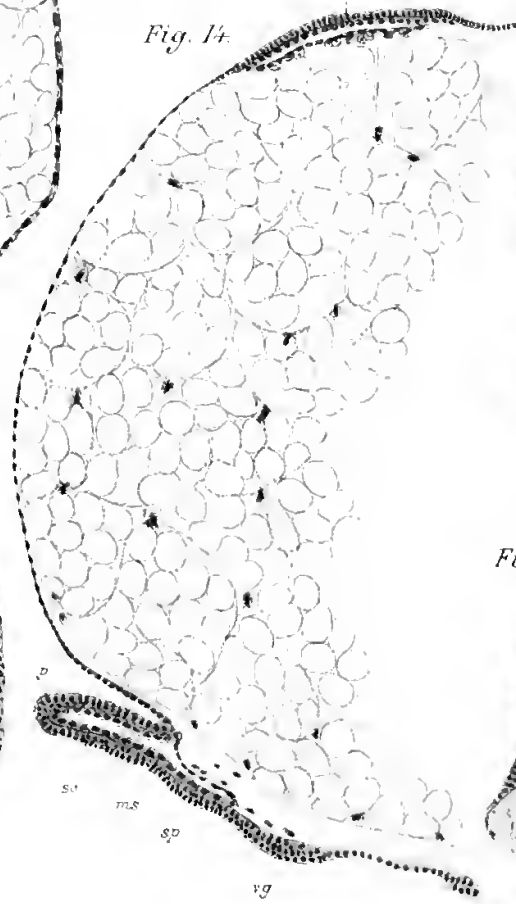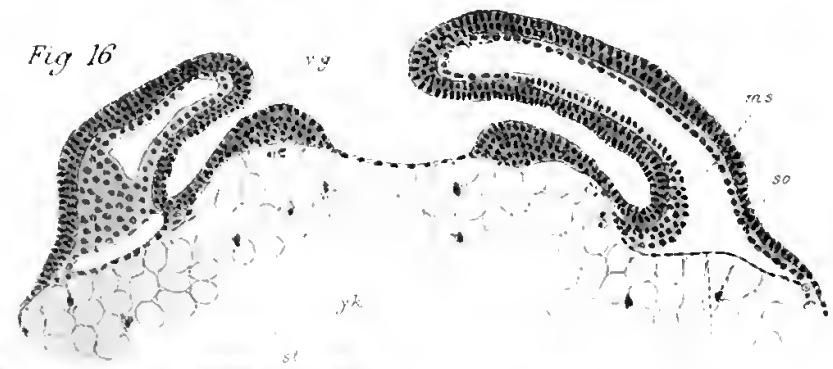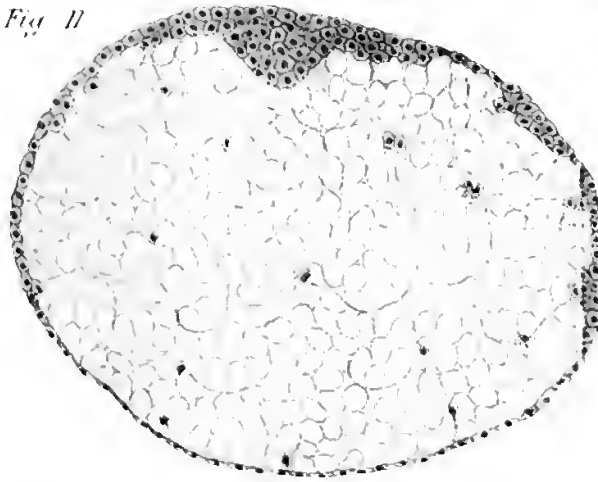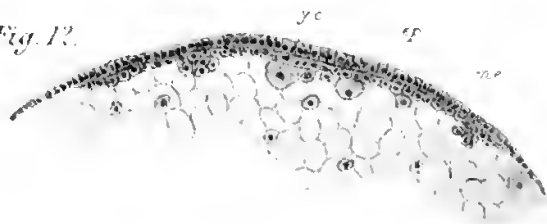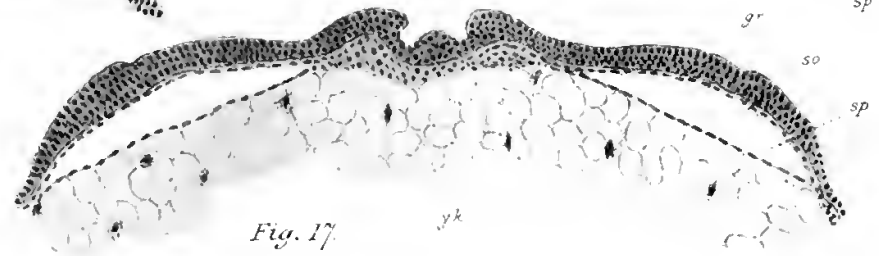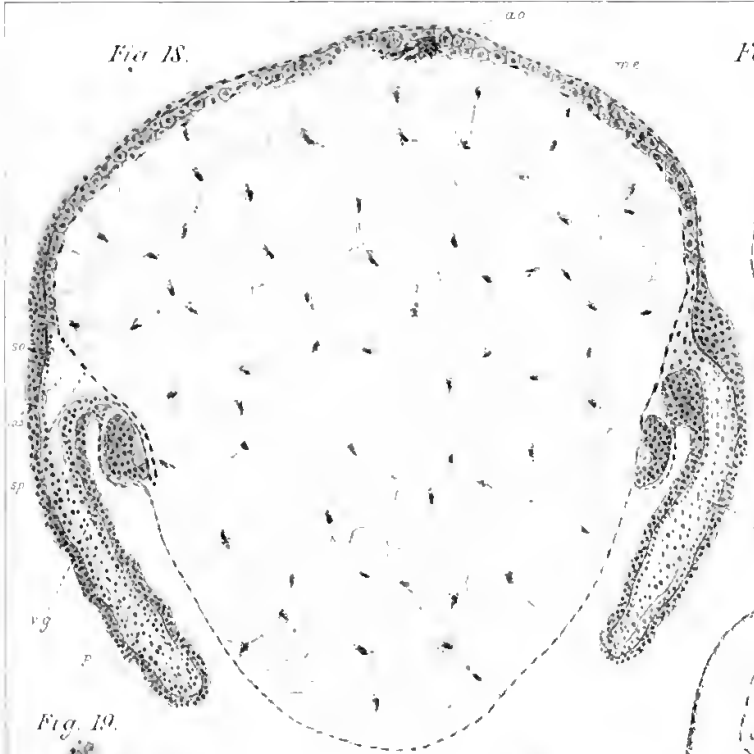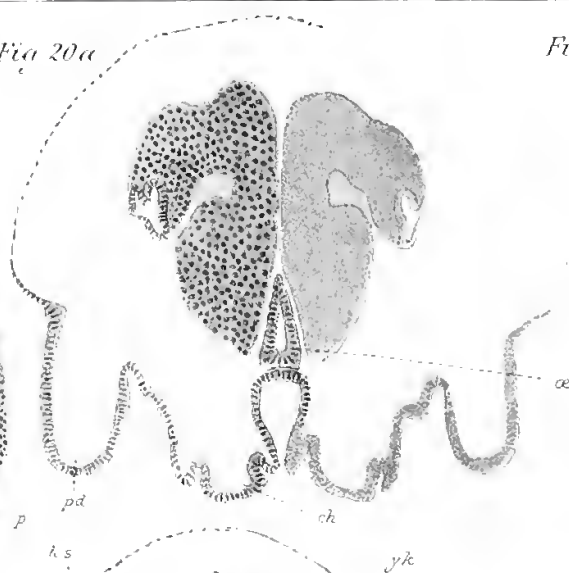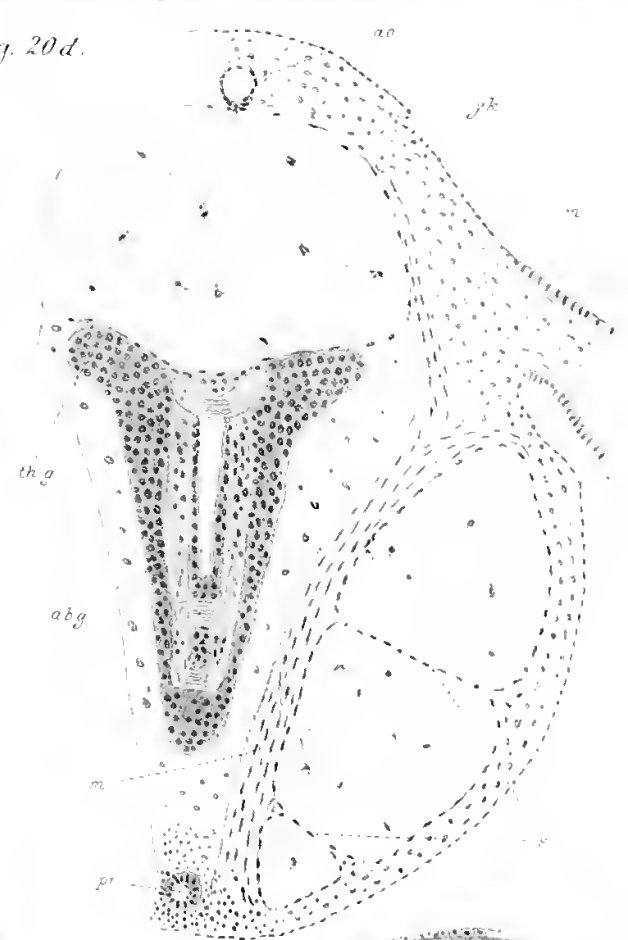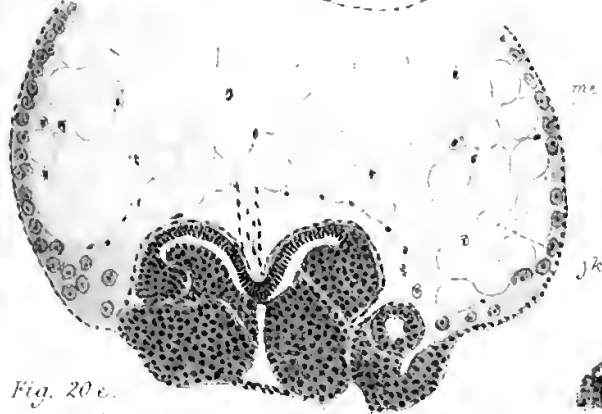
Fig. 12.

Fig. 13.

Fig. 14.

Fig. 15.

Fig. 16.

Fig. 17.
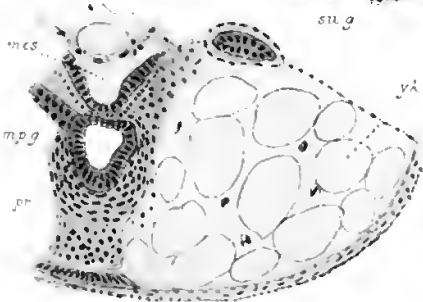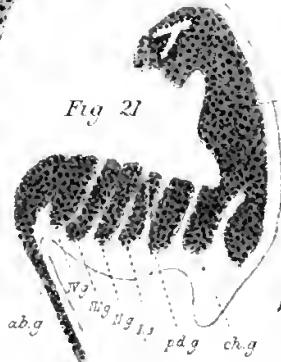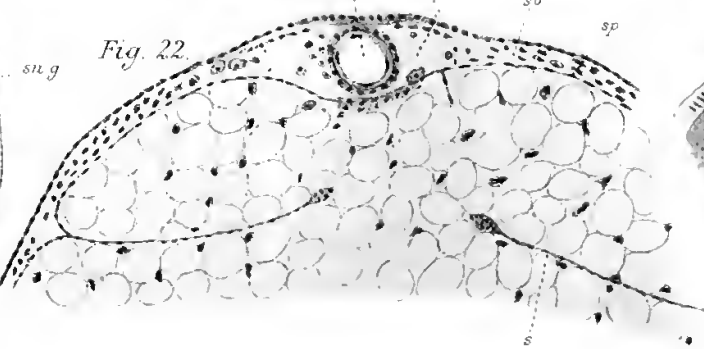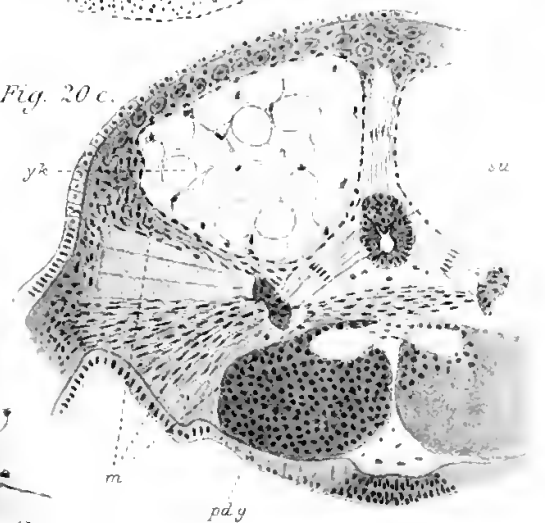
EXPLANATION OF PLATES XIX, XX, AND XXI,

Illustrating Mr. F. M. Balfour's Notes on the Development of the Araneina.

PLATE XIX.

Complete List of Reference Letters.

ch. g. Ganglion of cheliceræ. *c. l.* Caudal lobe. *ch.* Cheliceræ. *pd.* Pedipalpi. *pr. l.* Præoral lobe. *pp¹. pp².* *etc.* Provisional appendages. *p. c.* Primitive cumulus. *sp.* Spinnerets. *st.* Stomodæum.

I—IV. Ambulatory appendages.   1—16. Postoral segments.

Fig. 1.—Ovum, with primitive cumulus and streak proceeding from it.

Fig. 2.—Somewhat later stage, in which the primitive cumulus is still visible. Near the opposite end of the blastoderm is a white area, which is probably the rudiment of the procephalic lobe.

Fig. 3*a* and 3*b*.—View of an embryo from the ventral surface and from the side when six segments have become established.

Fig. 4.—View of an embryo, ideally unrolled, when the first rudiments of the appendages become visible.

Fig. 5.—Embryo ideally unrolled at the stage when all the appendages have become established.

Fig. 6.—Somewhat older stage, when the limbs begin to be jointed. Viewed from the side.

Fig. 7.—Later stage, viewed from the side.

Fig. 7*a*.—Same embryo as fig. 7, ideally unrolled.

Fig. 8*a* and 8*b*.—View from the ventral surface and from the side of an embryo, after the ventral flexure has considerably advanced.

Fig. 9.—Somewhat older embryo, viewed from the ventral surface.

PLATES XX AND XXI.

Complete List of Reference Letters.

ao. Aorta. *ab. g.* Abdominal nerve cord. *ch.* Cheliceræ. *ch. g.* Ganglion of cheliceræ. *ep.* Epiblast. *ht.* Heart. *hs.* Hemispherical lobe of supra-œsophageal ganglion. *l. l.* Lower lip. *m.* Muscles. *me.* Mesoblast. *mes.* Mesenteron. *mp. g.* Malpighian tube. *ms.* Mesoblastic somite. *œ.* Œsophagus. *p. c.* Pericardium. *pr.* Proctodæum (rectum). *pd.* Pedipalpi. *pd. g.* Ganglion of pedipalpi. *pr. c.* Primitive cumulus. *s.* Septum in abdomen. *so.* Somatopleure. *sp.* Splanchnopleure. *st.* Stomodæum. *su.* Suctorial apparatus. *su. g.* Supra-œsophageal ganglion. *th. g.* Thoracic ganglion. *v. g.* Ventral nerve cord. *yk.* Yolk. *y. c.* Cells derived from yolk. *y. n.* Nuclei of yolk cells.

I *g*—IV *g*. Ganglia of ambulatory limbs.   1—16. Postoral segments.

PLATE XX & XXI.—*Continued.*

Fig. 10.—Section through an ovum, slightly younger than fig. 1. Showing the primitive cumulus and the columnar character of the cells of one half of the blastoderm.

Fig. 11.—Section through an embryo of the same age as fig. 2. Showing the median thickening of the blastoderm.

Fig. 12.—Transverse section through the ventral plate of a somewhat older embryo. Showing the division of the ventral plate into epiblast and mesoblast.

Fig. 13.—Section through the ventral plate of an embryo of the same age as fig. 3, showing the division of the mesoblast of the ventral plate into two mesoblastic bands.

Fig. 14.—Transverse section through an embryo of the same age as fig. 5, passing through an abdominal segment above and a thoracic segment below.

Fig. 15—Longitudinal section slightly to one side of the middle line through an embryo of the same age.

Fig. 16.—Tranverse section through the ventral plate in the thoracic region of an embryo of the same age as fig. 7.

Fig. 17.—Transverse section through the procephalic lobes of an embryo of the same age. *gr.* Section of hemicircular groove in procephalic lobe.

Fig. 18.—Transverse section through the thoracic region of an embryo of the same age as fig. 8.

Fig. 19.—Section through the procephalic lobes of an embryo of the same age.

Fig. 20 *a, b, c, d, e.*—Five sections through an embryo of the same age as fig. 9. *a* and *b* are sections through the procephalic lobes, *c* through the front part of the thorax. *d* cuts transversely the posterior parts of the thorax, and longitudinally and horizontally the ventral surface of the abdomen. *e* cuts the posterior part of the abdomen longitudinally and horizontally, and shows the commencement of the mesenteron.

Fig. 21.—Longitudinal and vertical section of an embryo of the same age. The section passes somewhat to one side of the middle line, and shows the structure of the nervous system.

Fig. 22.—Transverse section through the dorsal part of the abdomen of an embryo of the same stage as fig. 9.
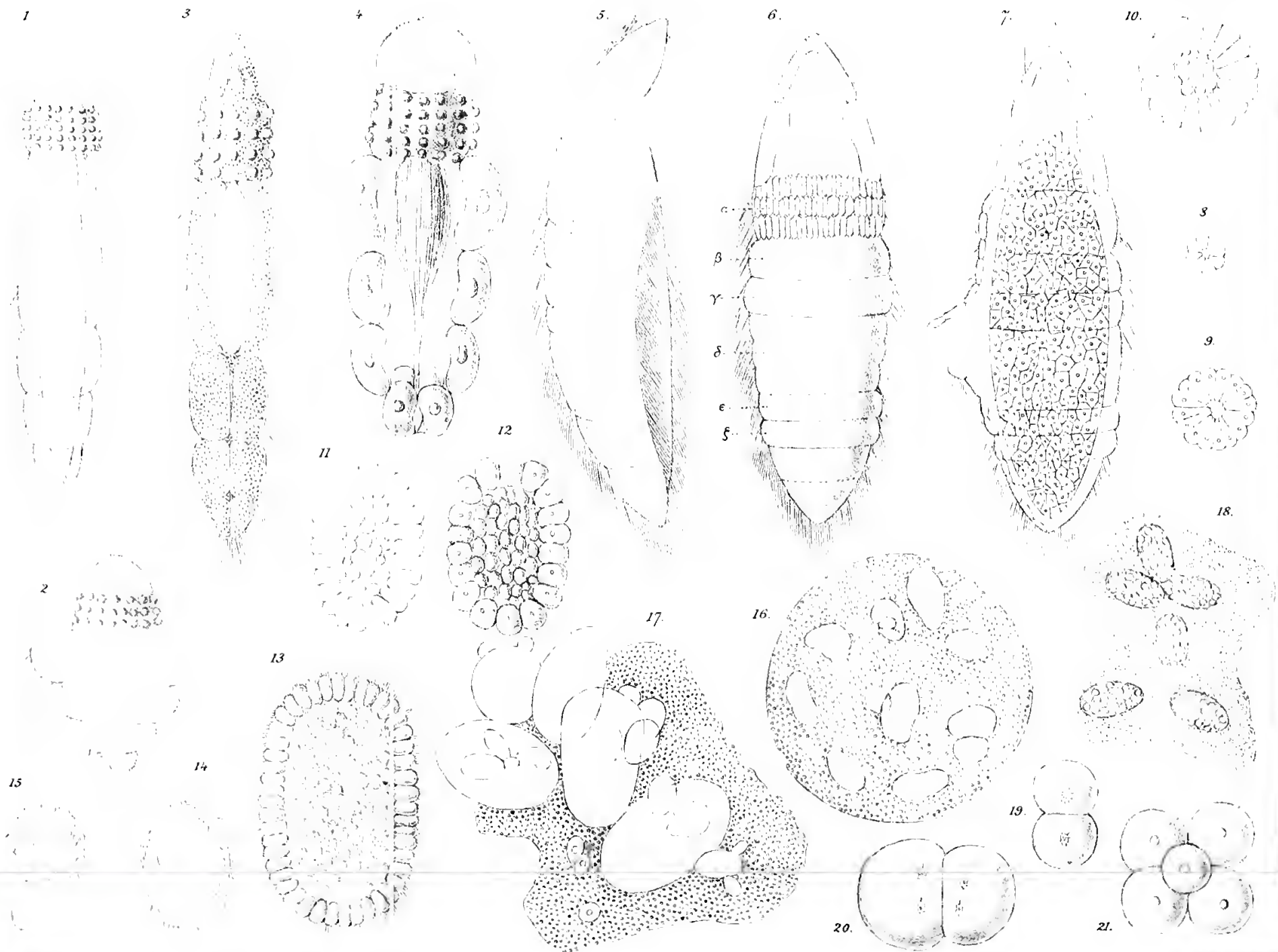
EXPLANATION OF PLATE XXII,

Illustrating Professor Giard's "Memoir on the Ortho-nectida."

Fig. 1.—*Rhopalura ophiocomæ* (natural state).

Fig. 2.—Short form, or young phase (natural state).

Fig. 3.—Adult animal, treated by reagents and showing the muscular bands.

Fig. 4.—Immature animal (acetic acid and carmine).

Fig. 5.—*Intoshia gigas* (profile view).

Fig. 6.—The same seen from the dorsal aspect, and treated in such a way as to show the ectodermal cells.

Fig. 7.—The same with the ectoderm removed, in order to show the endoderm.

Fig. 8.—Very young *blastula* of *Intoshia*.

Fig. 9.—The same more advanced.

Fig. 10.—Commencement of delamination.

Figs. 11, 12, and 13.—Formation of planula.

Figs. 14 and 15.—Very young sporocysts of *Intoshia* still retaining remains of the ectoderm.

Fig. 16.—More advanced sporocyst, opened in order to show its buds.

Fig. 17.—Portion of the same very highly magnified.

Fig. 18.—Bud in the condition of a *planula.*

Figs. 19, 20, and 21.—Stages in the segmentation of the egg of *Rhopalura*.
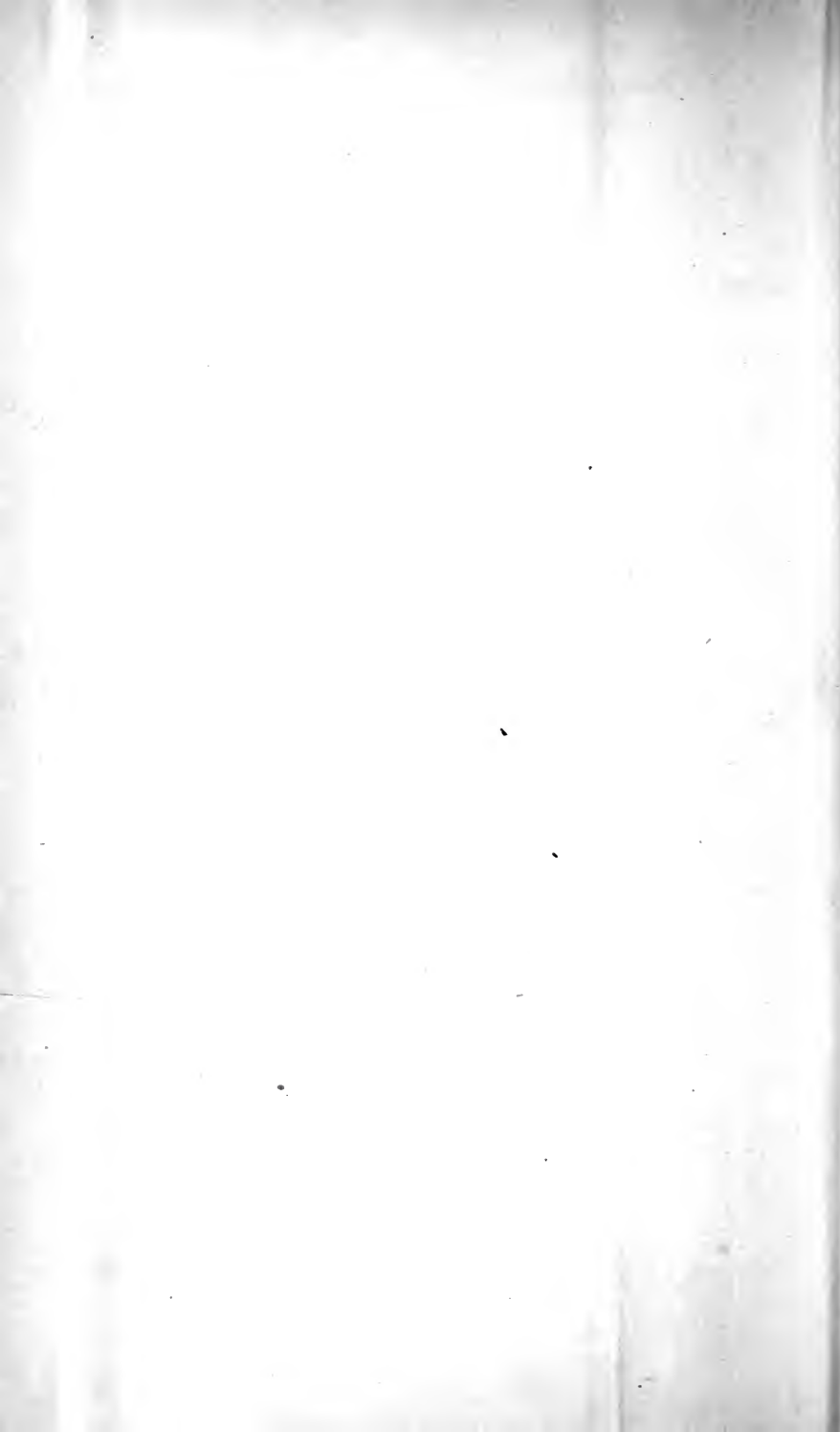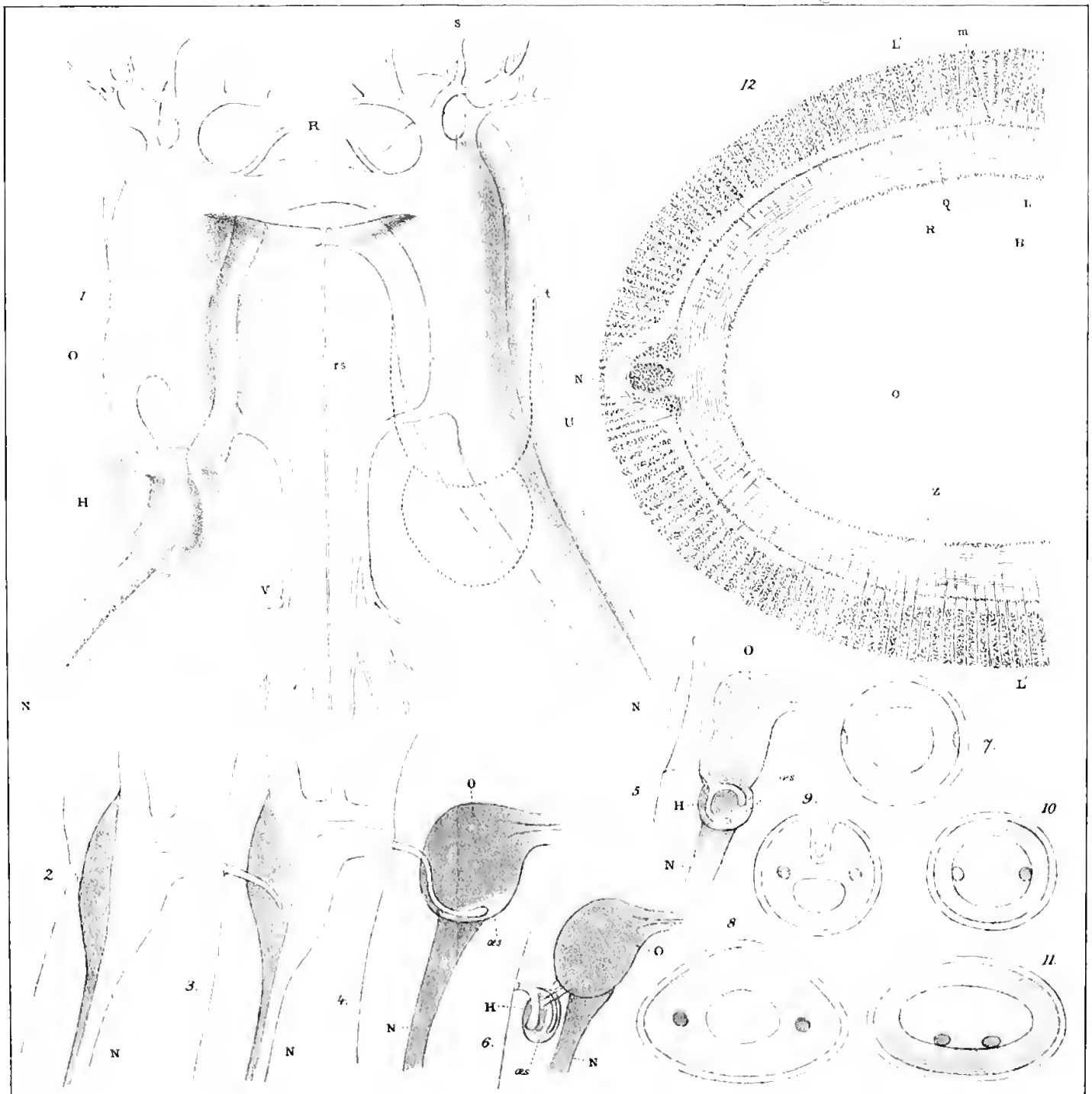
EXPLANATION OF PLATE XXIII,

Illustrating Dr. Hubrecht's "Researches on the Nervous System of the Nemertines."

Fig. 1.—The brain and the anterior portion of the lateral nerve-chords in *Cerebratulus,* and the LINEIDÆ in general (Diagrammatic).

An external sheath of nerve-cells is applied to an internal skeleton of nerve-fibre from which latter the principal nerves take their origin. In the figure the fibre is indicated by a lighter tint than the cells.

To the right the superior lobe (*O*) which is coalesced with the inferior lobe (*U*) anteriorly to *T* is supposed to have been removed. There is a thick ventral and a thin dorsal commissure. The median longitudinal nerve for the proboscidian sheath (*rs*) takes its origin out of the latter commissure.

H. Posterior or third brain lobe. *N.* Nerve-chords. *V.* Nervus vagus. *S.* Nervous stems to the tip of the snout, the eyes (when present) and the muscles of the lateral fissures.

Figs. 2—6.—Different stages of differentiation of the respiratory ciliated duct and of the mass of larger cells which are coalesced with the posterior brain lobe. This mass of cells is indicated by a lighter tint (in figs. 4, 5, and 6) than the mass of darker ganglion-cells. In figs. 2 and 3 the nerve-cells do not surround the nerve-fibre, but are applied externally to it; a mass of larger cells is not present here. The ciliated canal and its external opening are left white.

2. *Carinella annulata;* 3. *Carinella inexpectata;* 4. *Polia curta;* 5. *Cerebratulus roseus;* 6. *Drepanophorus.*

Figs. 7—11.—The different relative situations of the longitudinal nerve-chords in different genera. The epidermoidal tissues are left white, the muscles are darker and the nerve-chords of a darker shade still.

7. *Carinella;* 8. *Cerebratulus;* 9. *Langia;* 10. *Amphiporus;* 11. *Drepanophorus.*

Fig. 12.—Transverse section (left half) of the œsophageal region of *Cerebratulus roseus.* Neither the epiderm, nor the larger portion of the longitudinal muscles have been indicated.

O. Lumen of the œsophagus; *Z.* Folds in the mucous membrane; *Pr.* Proboscidian sheath with *B,* dorsal longitudinal blood-vessel; *L.* Thin inner layer of longitudinal muscles; *Q.* Circular muscular layer; *L'.* Exterior layer of longitudinal muscles; *M.* Median dorsal nerve-cord. *N.* Left nerve-chord with central fibrous portion separated from the ganglion cells by a hyaline sheath.

In direct continuous connection with the ganglion cells is a cellular sheath encircling the body and situated between the outer longitudinal and the circular muscular layers. It is this sheath which the author regards as a "nervous tunic."

EXPLANATION OF PLATES XXIV & XXV,

Illustrating Mr. A. G. Bourne's Memoir "On the Structure of the Nephridia of the Medicinal Leech."

References.

L. B. V. Lateral blood-vessels.   M. L. Cells of main lobe.   A. L. Cells of apical lobe.   T. L. Cells of testis lobe.   C. D. Central duct.   V. D. Vesicle duct.   R. D. Recurrent duct.

a. Nuclei.   *b.* Blood-vessels.   *b'.* Empty blood-vessels.   *c.* Cuticle of the blood-vessels.   *d.* Muscular fibres.   *d'.* Circular muscular fibres of lateral blood-vessels.   *d''.* Longitudinal.   *e.* Cells of salivary gland. *f.* Indifferent connective-tissue fibres.   *g.* Pigmented vaso-fibrous connective tissue.   *h.* Cilia in the vesicle.   *j.* Opening of vesicle duct into vesicle. *k.* Intracellular ductules.   *l.* Cuticle of ductules.   *m.* Cuticle of cells.

Fig. 1.—Semi-diagrammatic view of a segmental organ, taken from one of the right-hand series in the region of the testes. The blood-vessels are supposed to be injected, and the duct and ductules full. The connective tissue, which covers the organ and penetrates between the cells, and in which the blood-vessels lie, is not represented.

Fig. 2.—Termination of the apical lobe ("apex") and a portion of the testis lobe, showing the origin of the recurrent duct, R. D.

Fig. 3.—Surface view of cell from the apical lobe, with intracellular ductules, *k.*, fully distended with fluid.

Fig. 4.—Inner wall of the vesicle :—*j.* opening of the vesicle duct; *b.* blood-vessels ; *h.* cilia; V. D. vesicle duct ; *n.* ridges due to the partially contracted state of the vesicle.

Fig. 5.—Cells from the main lobe, with intracellular ductules fully distended.

Fig. 6.—Cells of the apical lobe, as seen in transverse section :— *b.* blood-vessels ; *c.* cuticle of blood-vessels ; *f.* indifferent connective-tissue fibres; *k.* intracellular ductules; *l.* cuticle of the ductules.   × 800 diams.

Fig. 7.—Cells of the main lobe seen in section, letters as before.

Fig. 8.—Cells surrounding recurrent duct in its free portion :—*a.* nucleus; *k.* intracellular ductules ; *l.* cuticle of the ductules ; *m.* cuticle of the cells.

Fig. 1.

Latero-abdominal Vessel.

Testis lobe.

Latero-abdominal Vessel.

Apical lobe.

Latero-dorsal Vessel.

Superior Vein.

Recurrent duct.

Main lobe.

Fig. 3.

Vesicleduct.

Vesicle.

Lateral Vessel.

Inferior Vein.

Fig. 4.

Fig. 8.

Fig. 5.

Fig. 7.

Fig. 6.

Fig. 2.

C.D

A.L

k

T.L

C.D

R.D   C.D   k

A. G. Bourne, del.

F. Huth, Lith. Edin.

Fig. 10.

Fig. 11

Fig 12

Fig. 9.

Fig 14

Fig. 13.
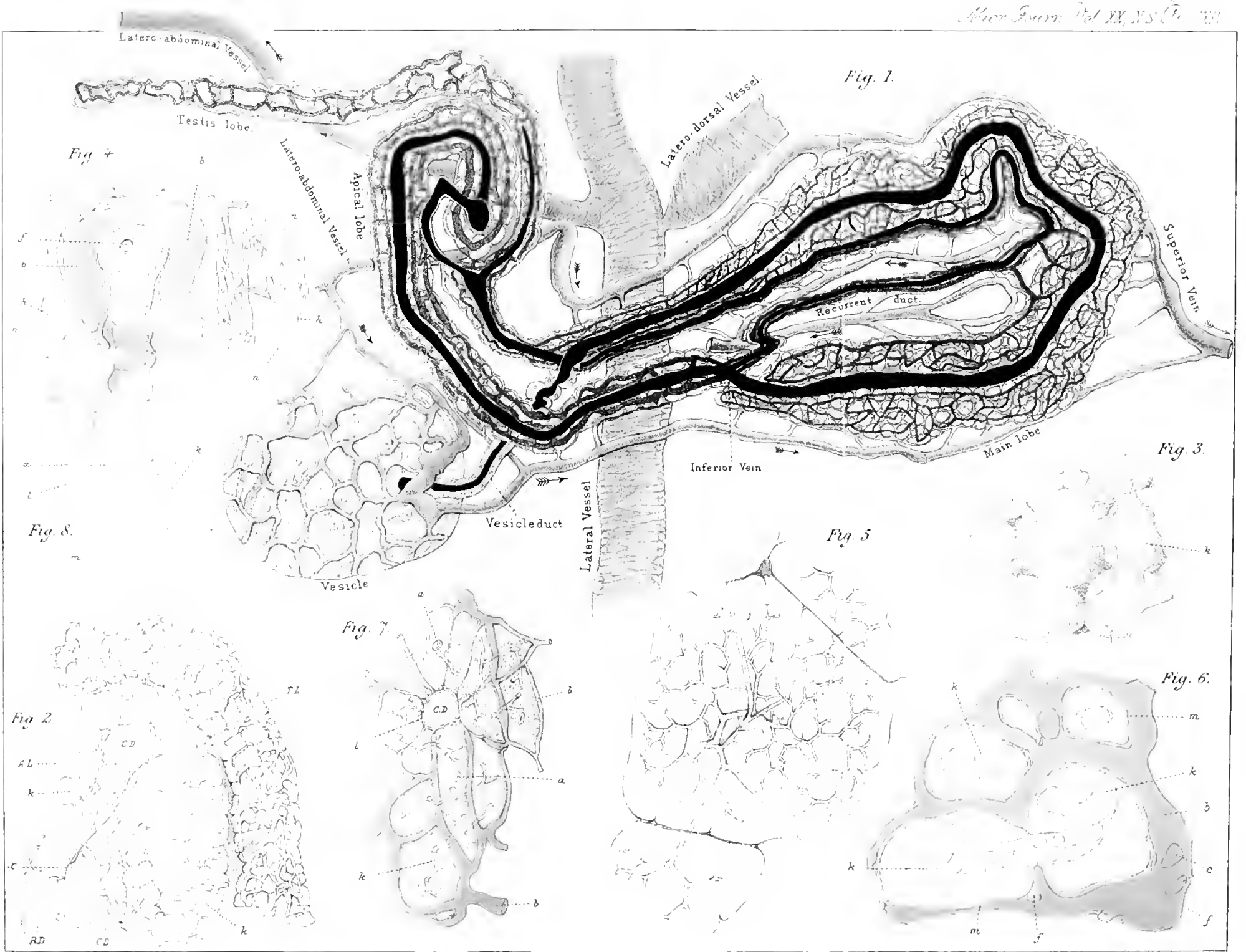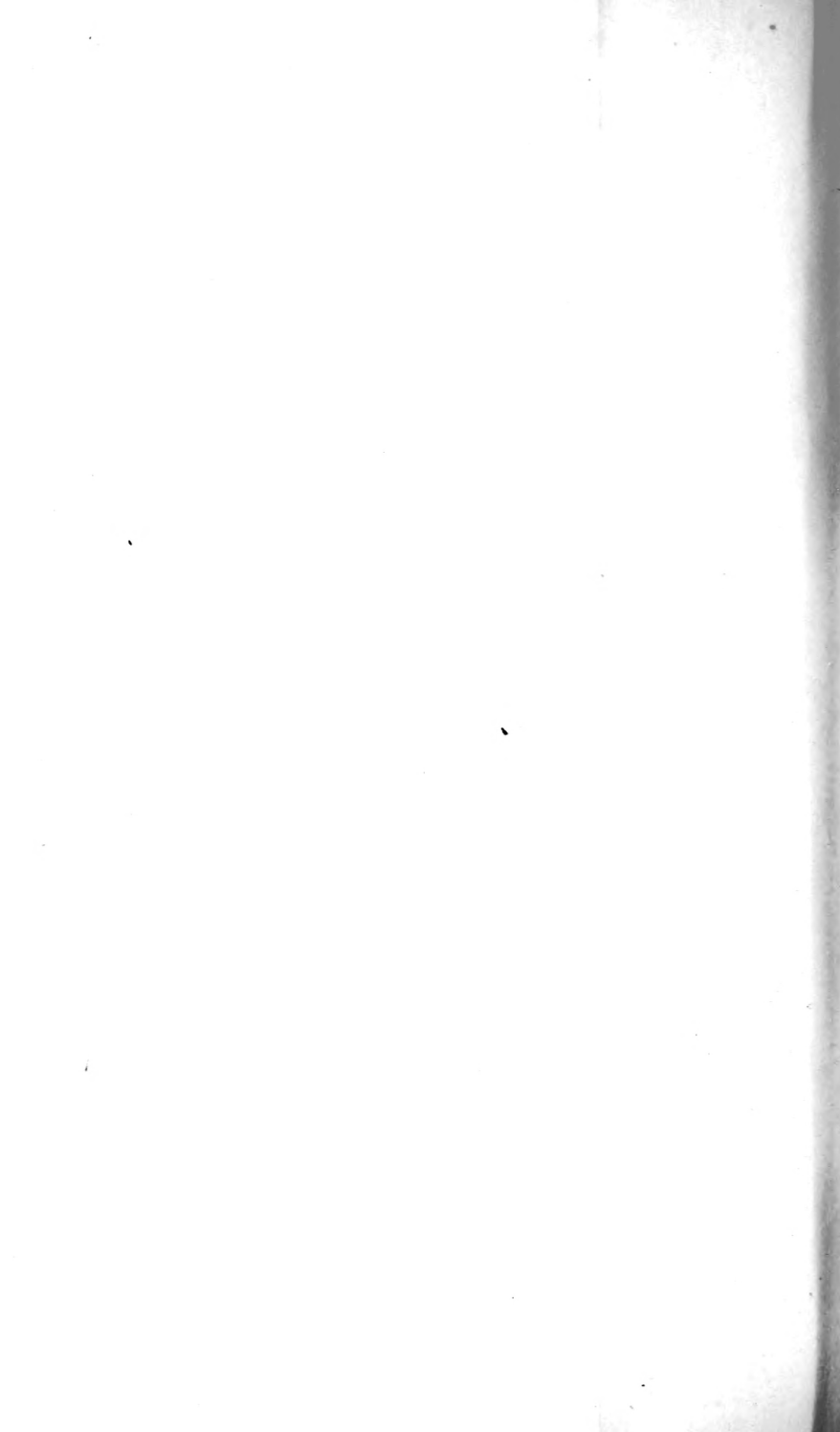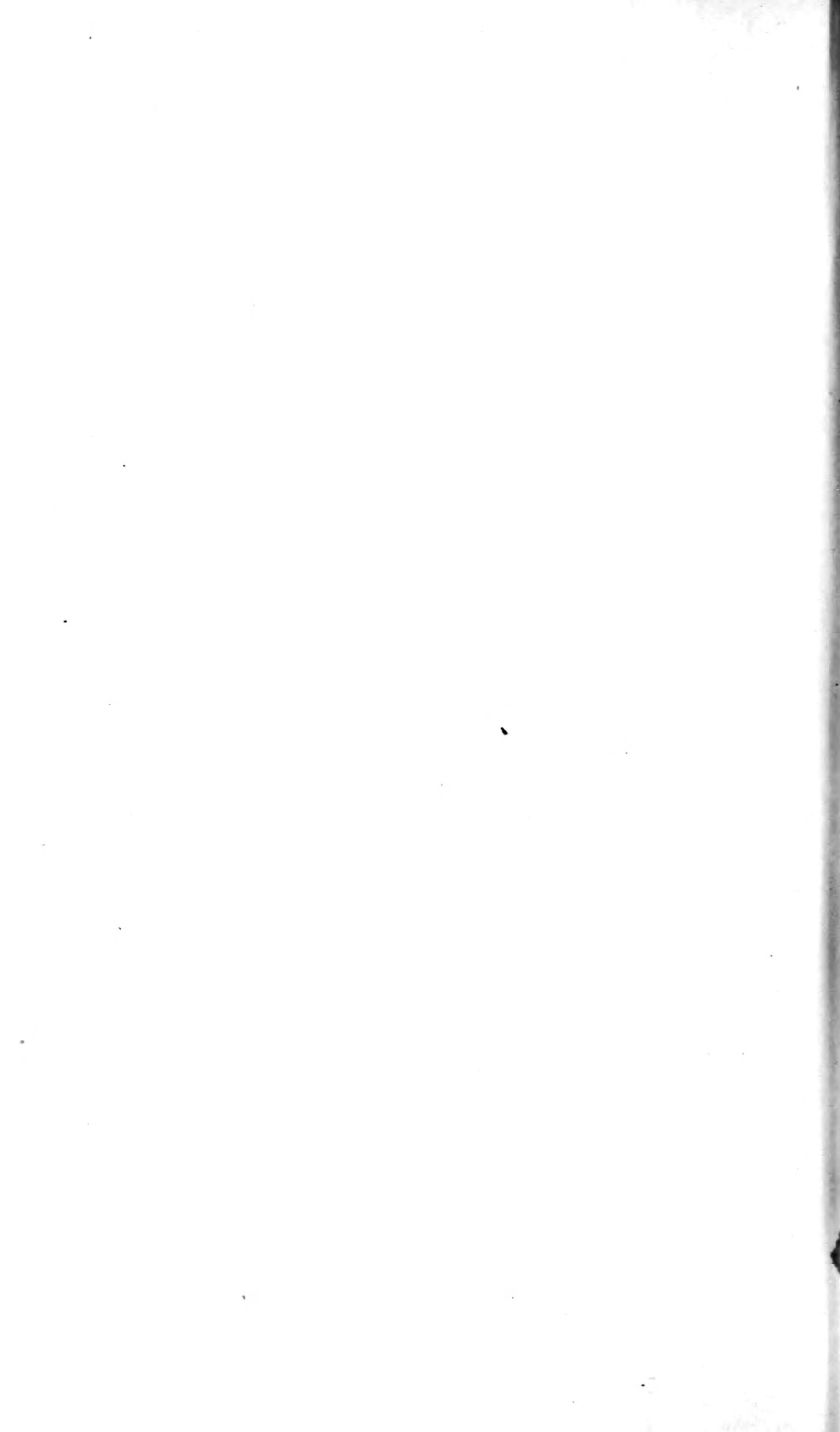
Fig. 9.—Surface view of cells from the apical lobe, after treatment with nitrate of silver.

Fig. 10.—Cells of the main lobe, after maceration in chromic acid $\frac{1}{10}$ per cent. :—*a.* nuclei, showing network ; *b.* blood-vessels ; *c.* cuticle of blood-vessels ; *f.* indifferent connective-tissue fibres ; *k.* intracellular ductules ; *l.* cuticle of the ductules ; *m.* cuticle of the cells.   × 800 diams.

Fig. 11.—Contents of the vesicle and central and recurrent ducts. × 3500 diams.

Fig. 12.—Contents of the ductules.   × 3500 diams.

Fig. 13.—Portion of a transverse section of a Leech :—L. B. V. lateral blood-vessel ; M. L. cells of the main lobe ; A. L. cells of the apical lobe ; T. L. cells of the testis lobe ; C. D. central duct ; V. D. vesicle duct ; R. D. recurrent duct.   *a.* nuclei ; *b.* blood-vessels ; *b'.* empty blood-vessels ; *d.* muscular fibres in longitudinal and transverse section ; *d'.* circular fibres of the lateral blood-vessel ; *d".* longitudinal fibres of the lateral blood-vessel ; *e.* cells of salivary gland ; *f.* indifferent connective-tissue fibres ; *g.* pigmented vaso-fibrous connective tissue ; *k.* intracellular ductules ; *l.* cuticle of the duct.   × 150 diams.

The nephridium here drawn lies on the left-hand side.   The cells of the main lobe are seen to lie above the lateral vessel, and the central duct has been cut across in three places in this region.   The central duct is seen again cut across twice, in the apical lobe, and the vesicle duct, which in the next section is free, is just becoming surrounded by the cells of the main lobe at the commencement of the lobe.   The recurrent duct, which further on lies in the cells of the main lobe, is here surrounded by testis lobe cells with their peculiar irregularly-shaped ductules.

Fig. 14.—Portion of a transverse section of a Leech, showing the vesicle and its excretory duct :—V. D. vesicle duct ; *b.* blood-vessels ; *d.* muscular fibres ; *f.* indifferent connective-tissue fibres ; *g.* pigmented vaso-fibrous connective tissue ; *h.* cilia.   × 150 diams.

EXPLANATION OF PLATE XXVI,

Illustrating Professor Ray Lankester's Memoir on "Intraepithelial Capillaries in the Integument of the Medicinal Leech."

Fig. 1.—Mallet-shaped cells of the tegumentary epithelium :—*b.* shows a portion of the cuticle adherent to the conjoined "heads" of two of the cells. Macerated preparation.

Fig. 2.—From a transverse section of a Leech, hardened in $\frac{1}{5}$ per cent. chromic acid, followed by alcohol :—*cu.* cuticle ; *v.* intra-epithelial blood-vessel ; *ep.* epithelial cells (mallet-shaped).

Fig. 3.—From a similar section :—*cu.* cuticle ; *v.* intra-epithelial vessel in transverse section ; *pg.* intra-epithelial pigment process (modified vaso-fibrous tissue) ; *ep.* mallet-shaped epithelial cells ; *gl.* superficial unicellular gland.

Fig. 4.—From a similar section :—*c. s.* compact connective substance (sub-epithelial layer of the integument). Other letters as in fig. 3.

Fig. 5.—Cuticle detached by maceration, showing openings of unicellular glands.

Fig. 6.—Surface view of a flake of tegumentary epithelium, prepared by maceration in potassium bichromate and staining with picro-carmine. The dark-coloured "handles" of the cells appearing like nuclei (and possibly representing such) are seen disposed in groups.

Figs. 7, 8, and 9.—Similarly prepared epithelial cells, showing the finely granular character of the cell-substance ; and in figs. 8 and 9, the perforation of the cell by the apertures of unicellular glands.

Fig 1.

Fig 2.

Fig. 8.

Fig 12

Fig 9.

Fig 10.

Fig 3.

Fig 4.

Fig 5.

Fig 11

Fig 7.

Fig 6.

Fig 13.

Fig 14.
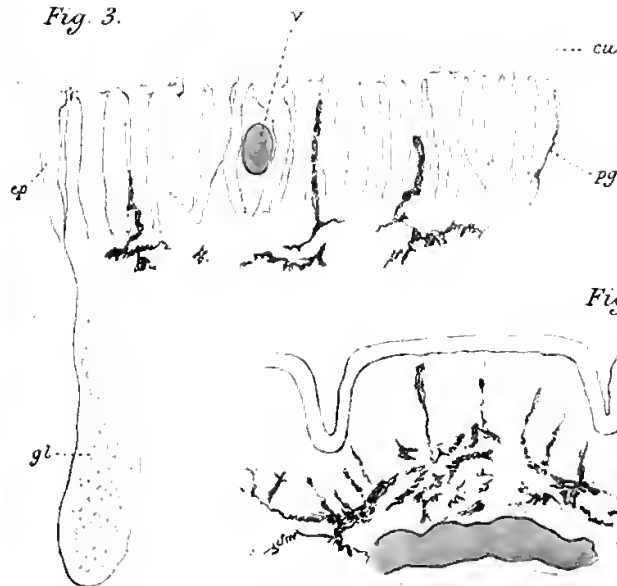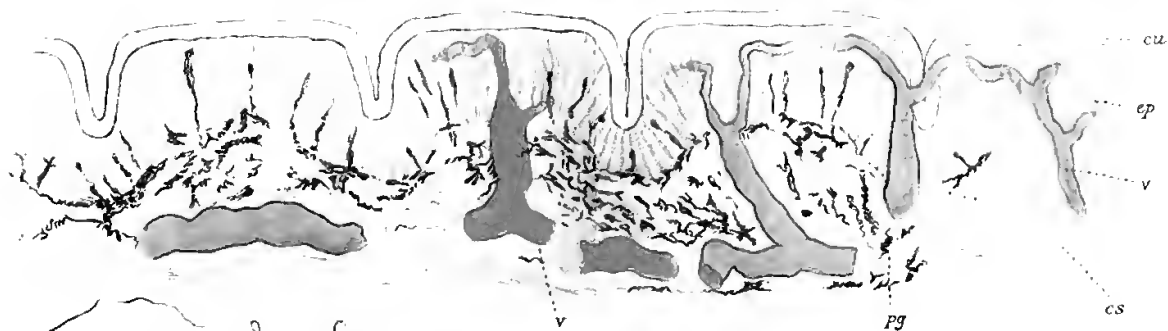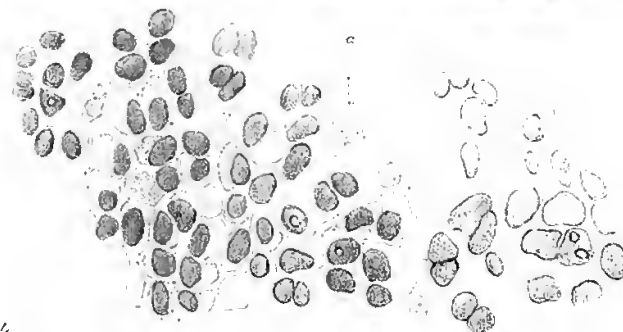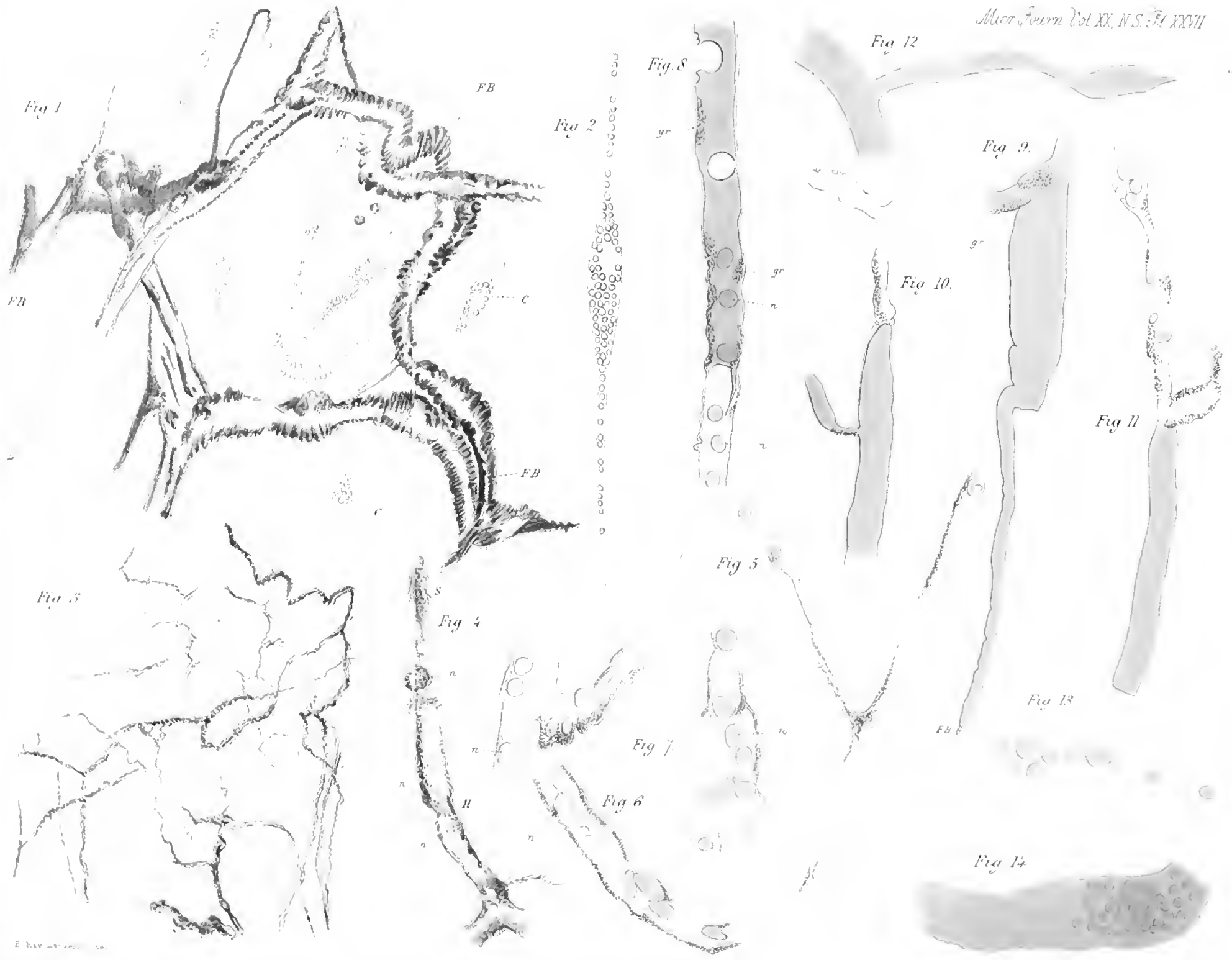
Fig. 17
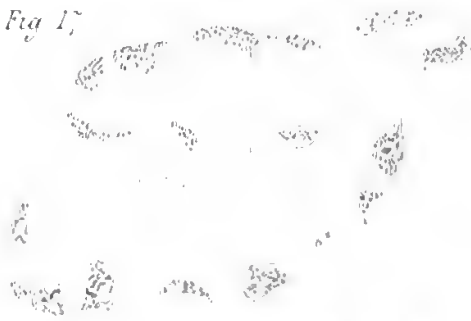
Fig. 15
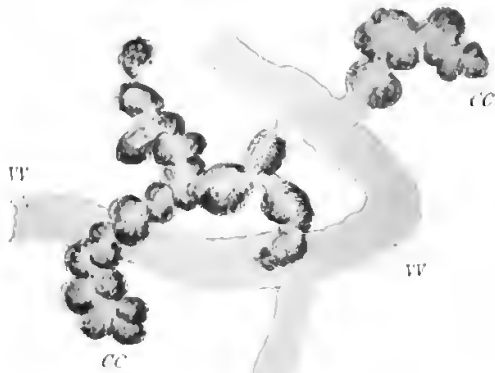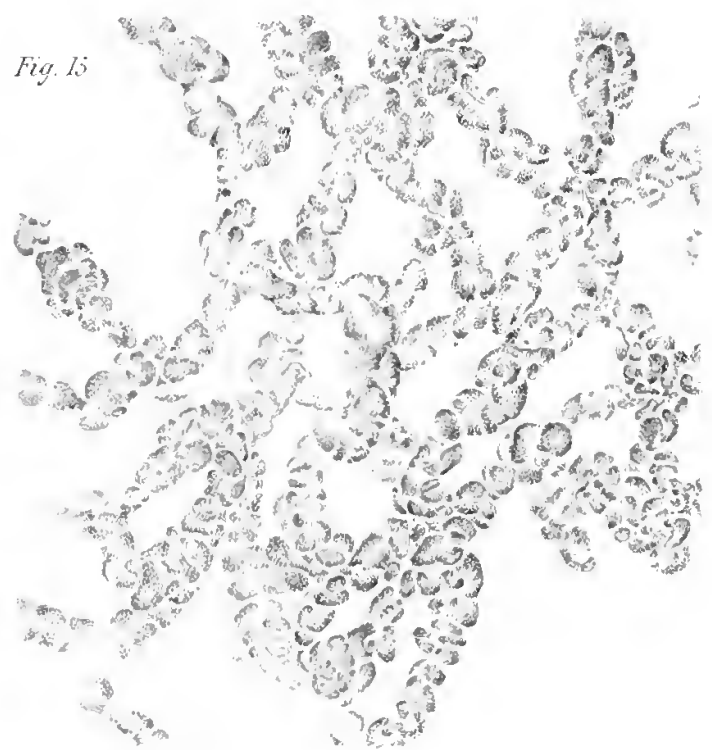
Fig. 18

Fig. 16
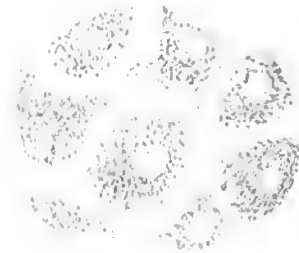
Fig. 19
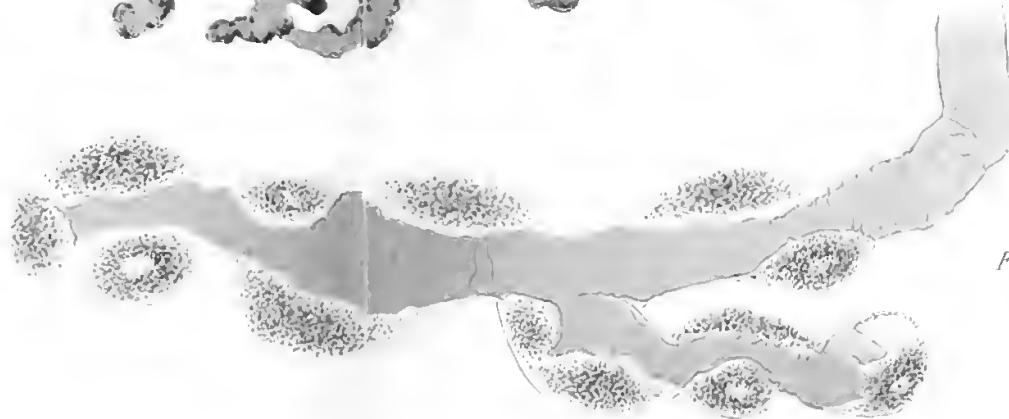
EXPLANATION OF PLATES XXVII AND XXVIII,

Illustrating Professor Ray Lankester's Memoir " On the Connective and Vasifactive Tissues of the Medicinal Leech."

Fig. 1.—Tissue from the neighbourhood of a nephridium, from a freshly-killed Leech. FB. Brown fibres of the entoplastic vaso-fibrous system. c. Branched and fusiform corpuscles of the ectoplastic connective jelly.

Fig. 2.—A single corpuscle of the connective jelly; nucleus stained by picro-carmine.

Fig. 3.—Network of brown fibres from the neighbourhood of the nerve-cord. Less magnified than fig. 1.

Fig. 4.—Brown fibre showing nuclei, n, and tubular character. S. Solid portion of the fibre. H. Hollow portion. Osmic picro-carmine preparation.

Fig. 5.—More decidedly tubular fibre of the vaso-fibrous system, showing in one part very thin wall devoid of granulations, and numerous nuclei projecting into the lumen of the tube. The diameter of nuclei is $\frac{1}{500}$th of an inch. Osmic picro-carmine preparation.

Figs. 6 and 7.—Similar tubular fibres, showing nuclei and granular walls.

Fig. 8.—Hæmatophorous vessel (blood-vessel) with structureless wall, distended with hæmoglobinous fluid, and continuous with a brown tubular fibre, in which are free nuclei. From a chromic-alcohol section, stained with picro-carmine.

Fig. 9.—Hæmatophorous vessel with structureless wall, showing a few granules at gr, and terminating at FB in a fibre.

Figs. 10 and 11.—Similar vessels to that drawn in fig. 8.

Fig. 12.—A similar vessel to that drawn in fig. 9.

Fig. 13.—Thin-walled hæmatophorous vessel containing free nuclei (corpuscles), stained pink by picro-carmine. The hæmoglobinous fluid has escaped.

Fig. 14.—A similar vessel, with smaller free nuclei—the hæmoglobinous fluid is here in position. Osmic picro-carmine preparation.

Fig. 15.—Botryoidal tissue of the medicinal Leech, drawn from a horizontal section of a chromic-alcohol preparation. I am indebted to Mr. W. E. Roth, Demonstrator in the Zoological Laboratory of University College, for this excellent drawing.

Fig. 16.—Surface view of the termination of one of the cæca of the botryoidal plexus in order to show the cells. Fresh preparation.

Fig. 17.—Transverse section of cæca of the botryoidal plexus in order to show the relation of the cells to the lumen of the vessel. The hæmoglobinous fluid was entirely absent in this portion of the preparation. Chromic-alcohol section.

Fig. 18.—Outlying portion of the botryoidal plexus in order to show the development of thick-walled botryoidal vessels (cc) as branches of a thin-walled hæmatophorous vessel (vv). Osmic teased preparation.

Fig. 19.—Cæcum of the botryoidal plexus in optical section, showing the coagulated hæmoglobinous fluid surrounded by the swollen granular cells of the vessel-wall.

Figs. 16, 17, and 19 are more highly magnified than figs. 15 and 18.
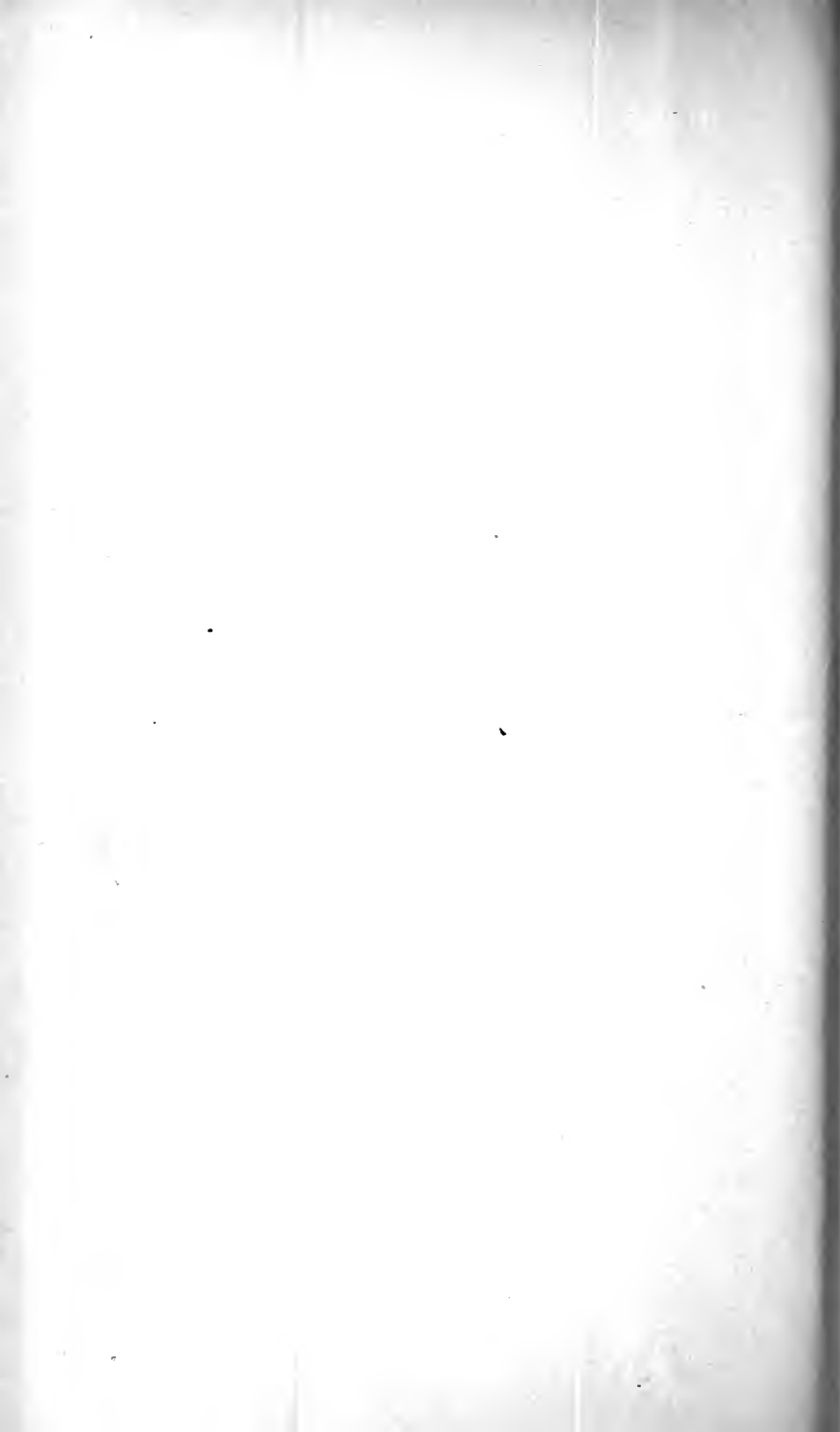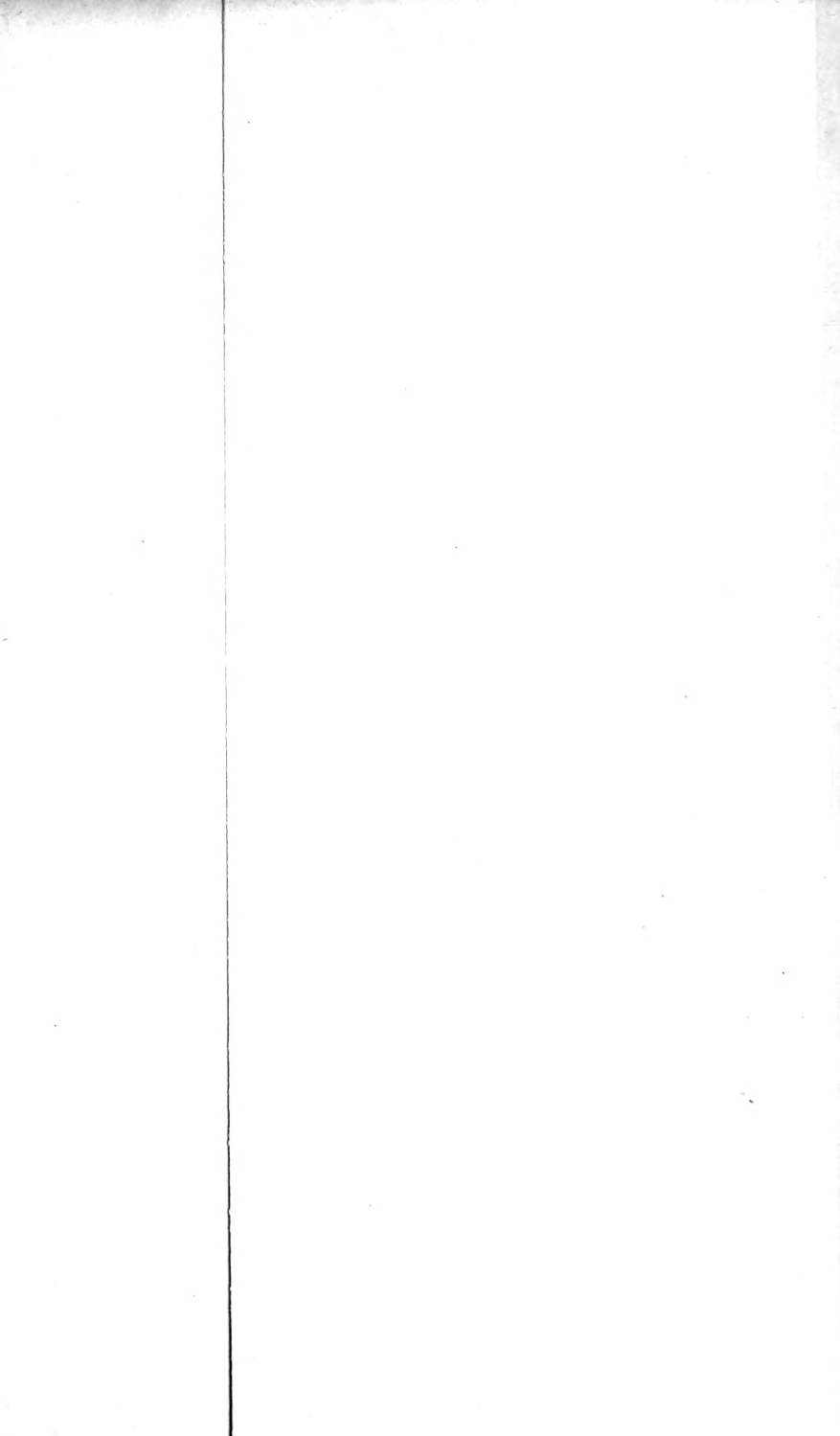
Fig 1

Fig 2

Fig 6.

Fig 3.
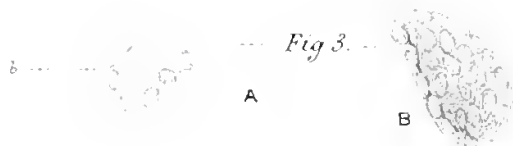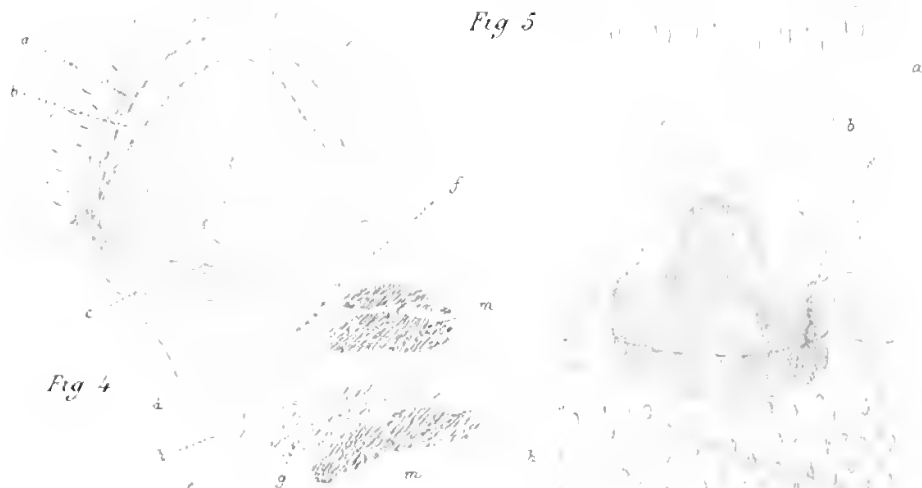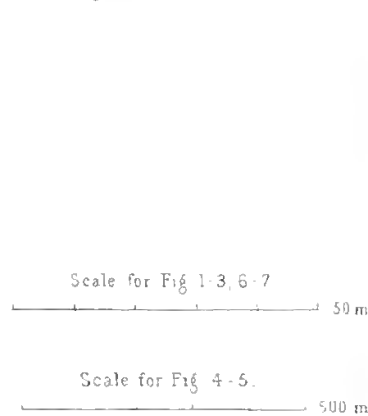
A

B

Fig 5

Fig 7

Fig 4

EXPLANATION OF PLATE XXIX,

Illustrating Professor Pouchet's Memoir " On the Laminar Tissue of Amphioxus."

Fig. 1.—Laminar network of the lophioderm.

Fig. 2.—Lophioderm from the region of the extremity of the notochord. *a*, cellular rods ; *c*, *c*, walls of the circular cavities with their cellular covering ; *c*, sheath of the notochord ; *d*, the largest cavity forming the transition to the cavities of the fin.

Fig. 3.—A. Section of a canal in the subdermic layer. *a*, dermis ; *b*, fibres. B. Pyriform cavity communicating with a cellular filament.

Fig. 4.—Transverse optical section of a papilla and its lumen. *a*, subcutaneous aponeurosis ; *b*, space filled with liquid surrounding the papilla, and completely lined with epithelium ; *c*, plan of the delicate fibres which send prolongations into the papilla ; *d*, *e*, amorphous substance covering each side of the median aponeurosis (*h*), and bounding on the other hand the blood (?) spaces, which are lined with cells ; *f*, *g*, amorphous substance in connection with the muscles (*mm*), and traversed by very large laminar fibres (*g*).

Fig. 5.—Longitudinal section passing through the median aponeurosis (*h*), on which are seen the nuclei of the cells, which cover the spaces shown in the preceding figure. The substance of the papilla presents concentric layers, which are clearly visible. The letters are the same as in the preceding figure.

Fig. 6.—Posterior extremity of the notochord, with the extremity of the central canal of the medulla, on which are inserted portions of the laminar network. In the tissue of the notochord are seen very small nuclei, which are very abundant and scattered throughout its whole thickness.

Fig. 7.—Sensory nerve terminations of the Trigeminus. *a*, simple termination with three cells, furnished with an envelope having itself a nucleus ; *b*, compound terminations.

Fig 2 Fig 1 Fig 4

Fig 2

Fig 1

Fig 4

Fig 8

Fig 5

Fig 3

Fig 7

Fig 9

Fig 6

LIMNOCODIUM SOWERBII.

Fig 10

Fig 11

Fig 14

Fig 12

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Fig 16

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Fig 17

Fig 19

Fig 18

Fig 20
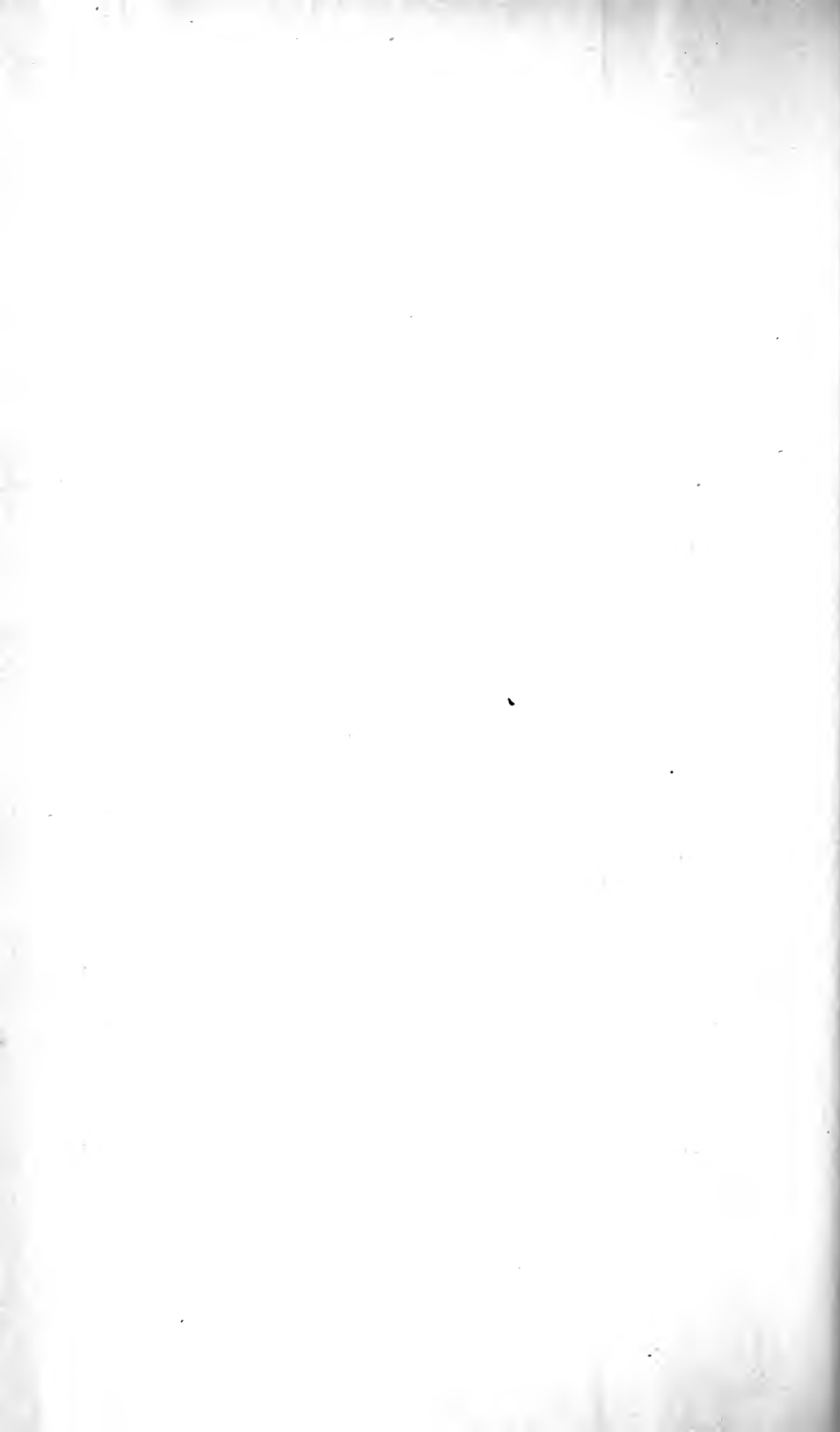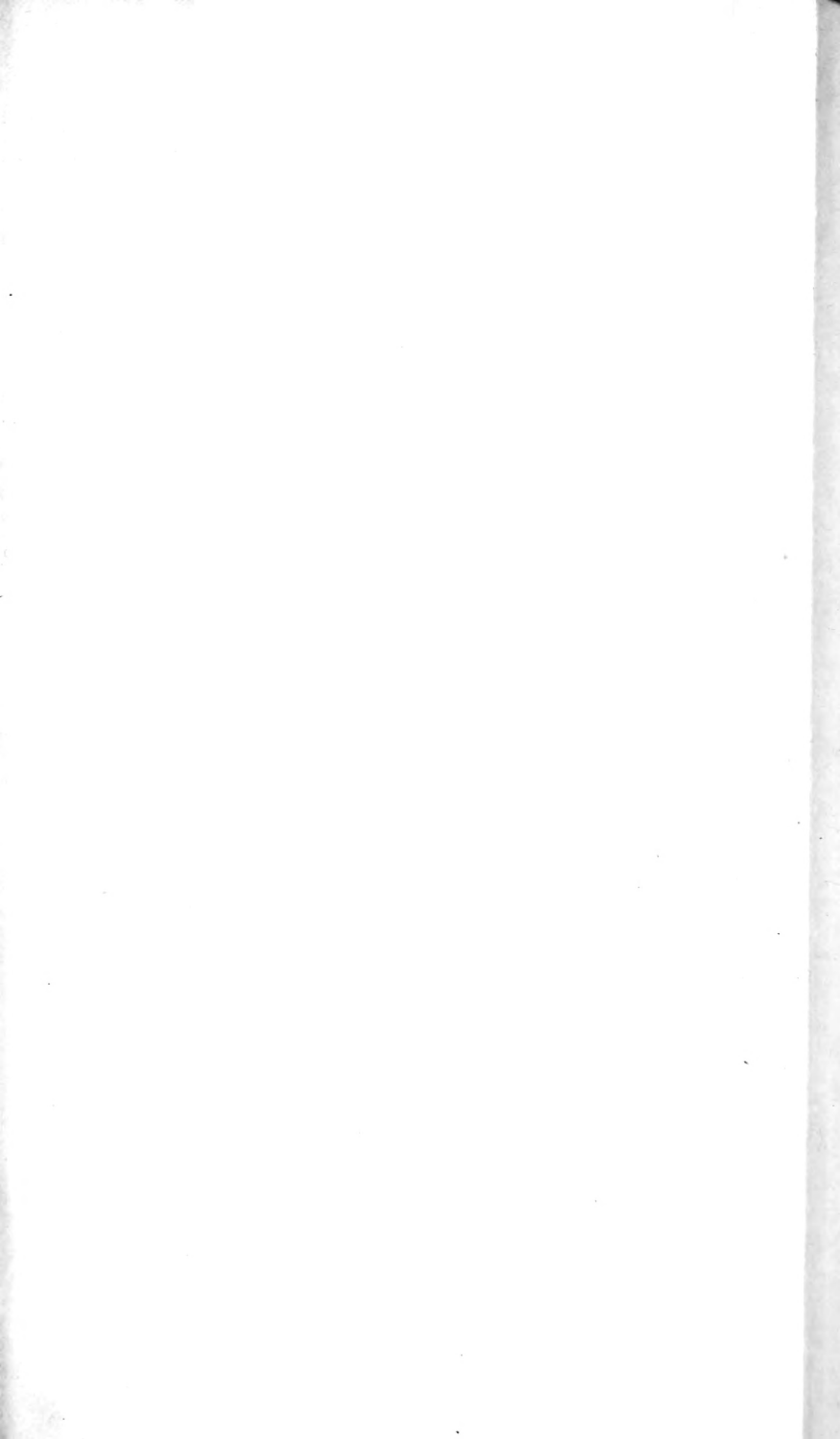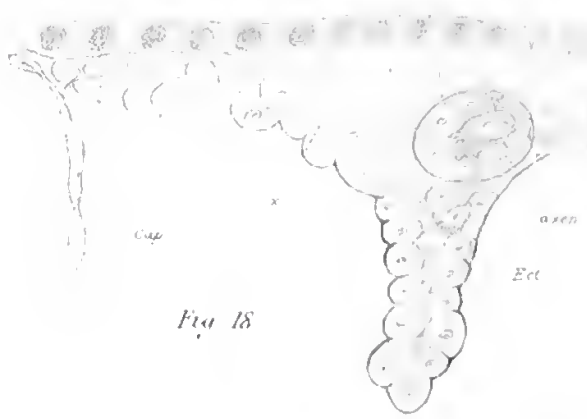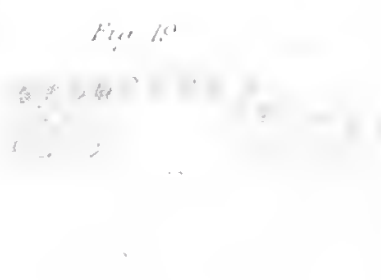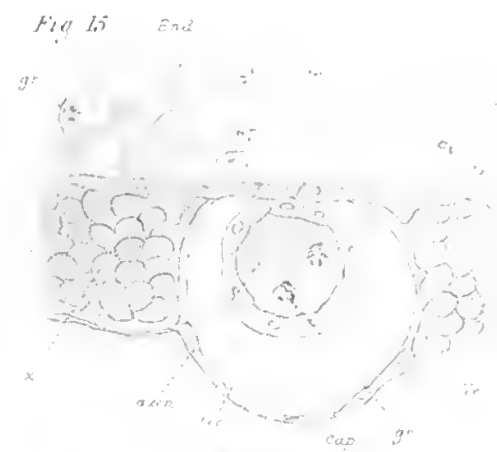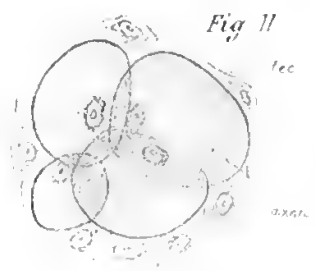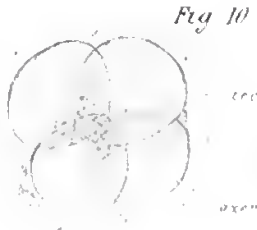
LIMNOCODIUM SOWERBII

EXPLANATION OF PLATES XXX AND XXXI,

Illustrating Professor Ray Lankester's Memoir on "*Limnocodium* (Craspedacustes) *Sowerbii*, a new Trachomedusa inhabiting fresh water."

Fig. 1.—Enlarged sketch of a quadrant of the disc of an immature specimen ($\frac{1}{3}$ of an inch in diameter) seen from the subumbral surface. *Per T.* two of the perradial tentacles; *Ax.* endodermal axis of same; *MR.* marginal ring (nettle-ring, peronia, and endodermal ring tissue); *MC.* marginal canal; *RC.* radial canals; *VMC.* velo-marginal cells; *Ve.* velum in which are seen the "velar centrifugal canals" or tubular capsules of the marginal bodies. Just above *Ve.* two neighbouring capsules are seen to have coalesced; *Go.* the immature outgrowths of the radial canals, in the ectoderm of which the generative products are formed; *St.* stomach.

Fig. 2.—Edge of the velum as seen when reflected, showing the position of a tubular "otocystic" capsule between the two layers of the velum; *ot.* tubular capsule; *Sup.* abumbral ectoderm-layer of the velum; *Inf.* the adumbral musculo-epithelial ectoderm of the velum; *Vac.* vacuolated cells of the abumbral ectoderm, which by fusion cause the increase of the tubular capsule; *musc.* circular muscle-fibres of the adumbral ectoderm (not shown in the lithograph).

Fig. 3.—Diagram of a section through the edge of the disc to show the relation of the tentacle-root to the endoderm of the marginal canal; *Ec T.* ectoderm of the tentacle; *En T.* endoderm of the tentacle; *Ec D.* supraumbral ectoderm of the disc; *Ec D'.* subumbral ectoderm of the disc; *Ec V.* abumbral ectoderm of the velum; *Ec V'.* adumbral ectoderm of the velum; *NR.* nettle-ring, viz. ectoderm of the marginal ring charged with thread-cells; *En R.* endoderm of the abumbral wall of the marginal canal, having a cartilaginoid character and greenish-yellow colour; *VMC.* one of the strongly marked row or ridge of endodermal cells termed "velo-marginal cells;" *En L.* position of the endoderm-lamella of the disc; *x.* ring of colourless ectodermal cells, being the abumbral portion of the nerve-ring; *Li.* marginal body; *Cap.* tubular capsule of the marginal body; *ot.* cavity of the tubular capsule; *MC.* marginal canal.

Fig. 4.—View from the supra-umbral surface of a portion of the marginal ring; the ectodermal cells only are drawn. *T.* tentacle; *TR.* tentacle-root; *Ec D.* ectoderm of the supra-umbral surface of the disc; *Pe.* peronium; *NR.* nettle-ring; *Ec V.* ectoderm of the abumbral surface of the velum; *VMC.* row of velo-marginal cells (endodermal) showing through the ectodermal cells of the nettle-ring.

Fig. 5.—The same preparation more deeply focussed. *T R.* tentacle-roots formed by notochordal tissue; *R End.* cartilaginoid endoderm of the marginal canal (representing the "ring-cartilage"); *MCa.* adumbral wall of the marginal canal in section, showing soft ciliated cells with contained granules (enlarged in fig. 7); *Mcp.* the inferior border of the marginal canal formed by the velo-marginal cells, marked *VMC* in other figures; *X.* nerve-ring; *Ec V.* adumbral ectoderm of velum; *MB.* marginal body; *CC.* tubular capsule of the same.

Fig. 6.—Transition of the pale notochordal tissue of a tentacle-root into

the green-coloured cartilaginoid tissue of the abumbral wall of the marginal canal.

Fig. 7.—Two cells of the adumbral wall of the marginal canal, showing their rounded form and dark greenish-brown-coloured concretionary granules.

Fig. 8.—One of the marginal bodies as seen in the living condition. The attached pole is the lower in the figure. A greenish granule similar to those of the endoderm cells is seen in the body to the left.

Fig. 9.—A similar body after the action of dilute acetic acid. Surface view.

Figs. 10 and 11.—Two marginal bodies or "refringent sense-bulbs" seen in optical section after the action of osmic acid. *tec.* cortical cells or tentacle-ectoderm; *axen.* medullary cells or endodermal axis.

Figs. 12 and 13.—Earliest stage in the development of a "marginal body" from a medusa nearly half an inch in diameter. *End.* endoderm of the marginal ring with green-coloured cell-substance; *Ect.* colourless ectoderm; *axen,* axial cell of the marginal body derived from endoderm and coloured by green particles; *cc.* tubular capsules of neighbouring "bodies."

Fig. 14.—Later stage in the development of the marginal body and its tubular capsule; *End.* endoderm of the marginal ring; *axen.* axial endoderm cells of the "body"; *Ect.* ectoderm of the velum; *tec.* tentacular ectoderm or cortical cells of the marginal body; *cc.* cavity of the tubular capsule.

Fig. 15—A later stage. *End.* endoderm of the marginal ring; *x.* nervering; *Ve.* velum; *gr.* greenish granules characteristic of the endoderm; *axen.* axial endoderm or medullary cells of the marginal body; *tec.* cortical cells or tentacular ectoderm of the same; *Cap.* tubular capsule.

Fig. 16.—A still later stage; *axen.* axial endoderm or medullary cells of the marginal body; *tec.* tentacular ectoderm or cortical cells of the marginal body; *cc.* cavity of the ectodermal tubular capsule; *cap.* wall of the capsule formed by the velar ectoderm.

Fig. 17.—Early stage in the development of a marginal body, in which the form of a tentacle is abnormally assumed; *End.* endoderm of the marginal canal; *Ect.* ectoderm of the abumbral velar surface; *axen.* enlarged axial endoderm cell, with green granules.

Fig. 18.—Abnormally developed marginal body devoid of capsule and having the form of a small free tentacle; *End.* endoderm; *Cap.* capsule of a neighbouring marginal body; *axen.* axial endoderm cells forming a spherical group; *ect.* tentacular ectoderm here having the form of a tentacle instead of closely investing the axial endoderm-sphere.

Fig. 19.—Abnormal capsule developing without any tentacular element, that is, without a marginal body; *End.* endoderm; *cc.* cavity of the capsule; *Cap.* wall of the capsule; *x.* nerve-ring.

Fig. 20.—Abnormal marginal body (seen in optical section) which has broken away from its attachment to the marginal ring and is lying free in the distal end of its tubular capsule. The body has taken on the form of a spherical cyst, and shows the cortical cells forming the wall of the cyst, whilst within project the large endodermal cells.
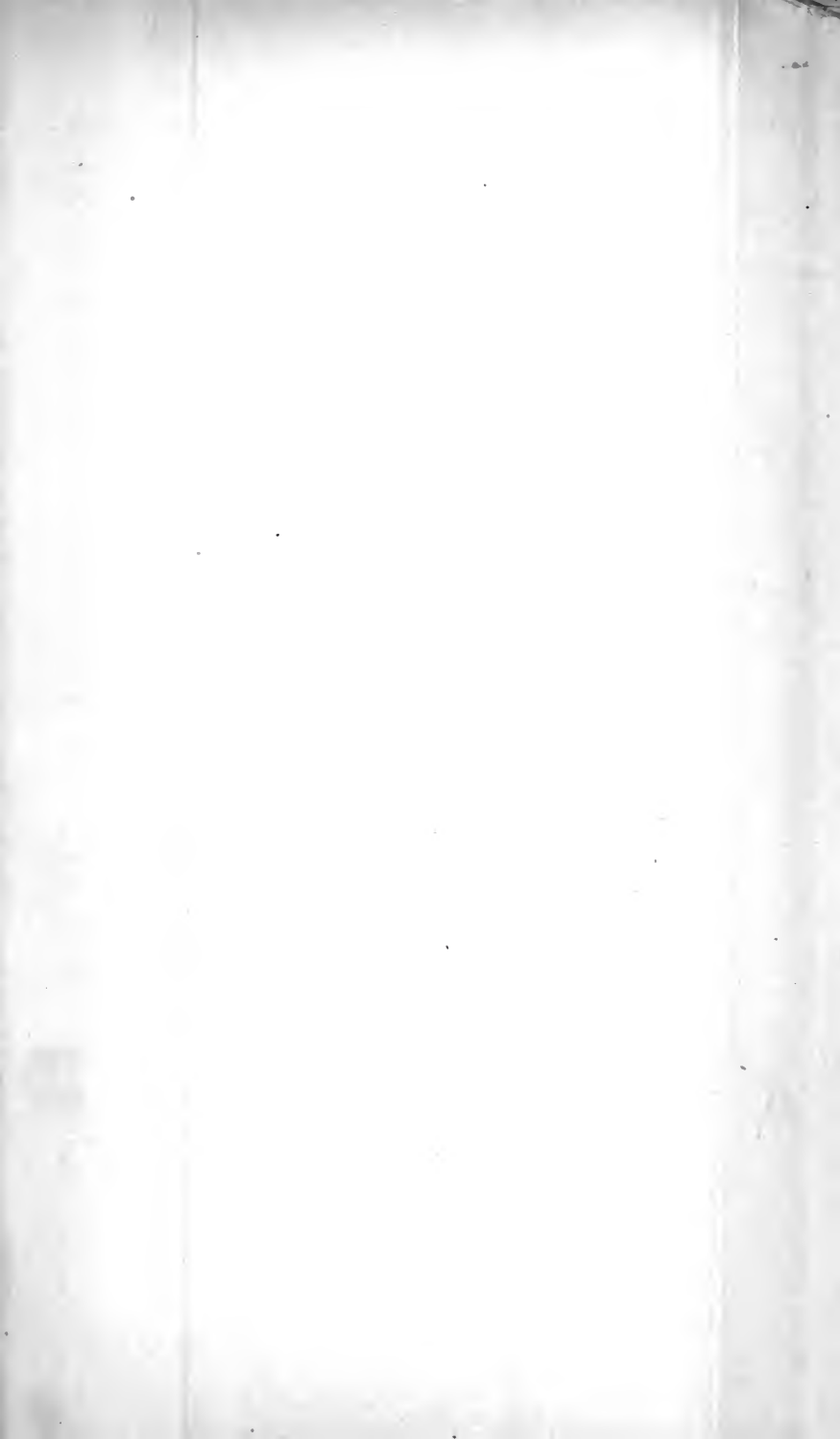
Fig 1 Fig 2 Fig 3 Fig. 5. Fig 7
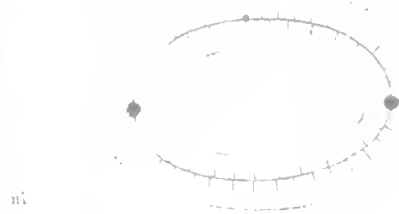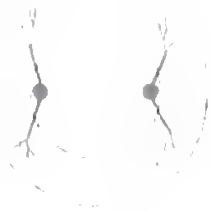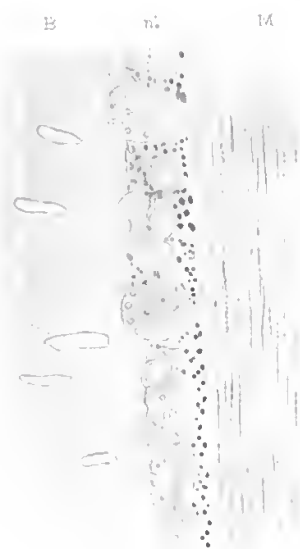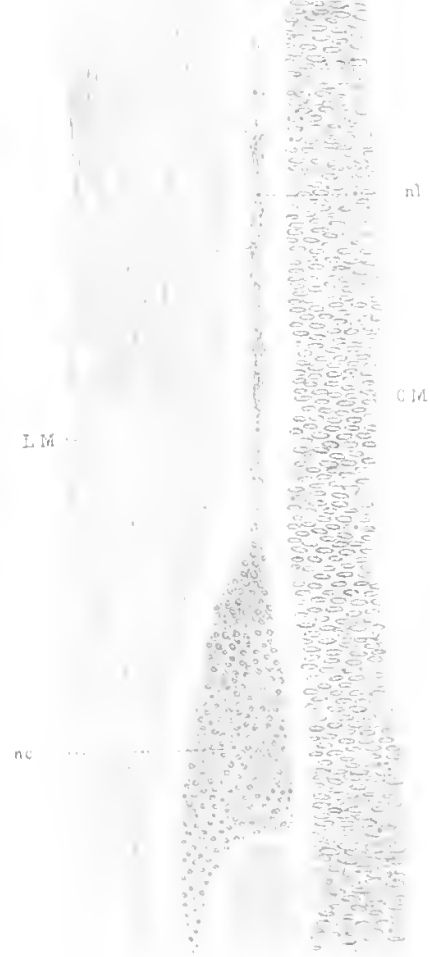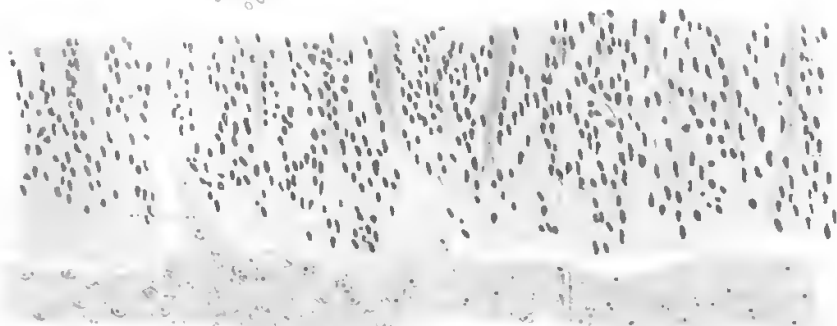
Fig 6

Fig. 4

Fig 8

Fig. 9.

Fig. 12

nps

LM

nc

B

rf

LM

nl

P
P

rf

IL

TM

nl CM

Fig. 11.

Fig. 10

LM

B

nl

l

nc

nps

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IL

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LM

LM

Fig. 13

nc

nc

N

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r'
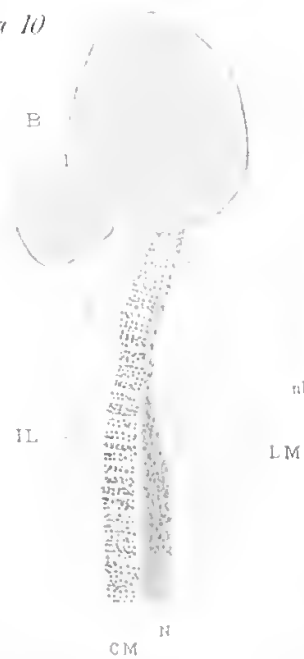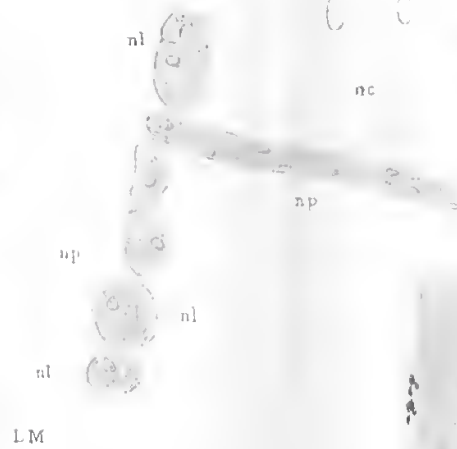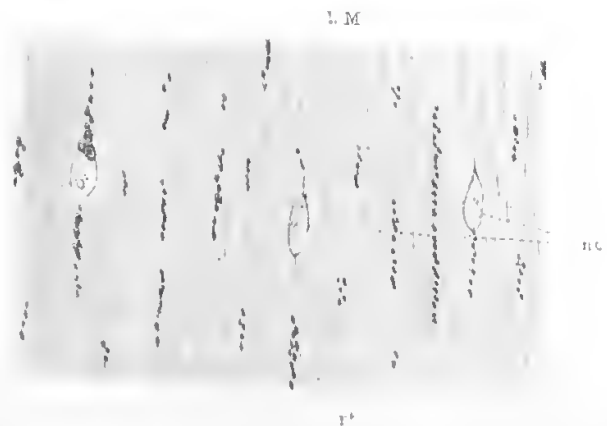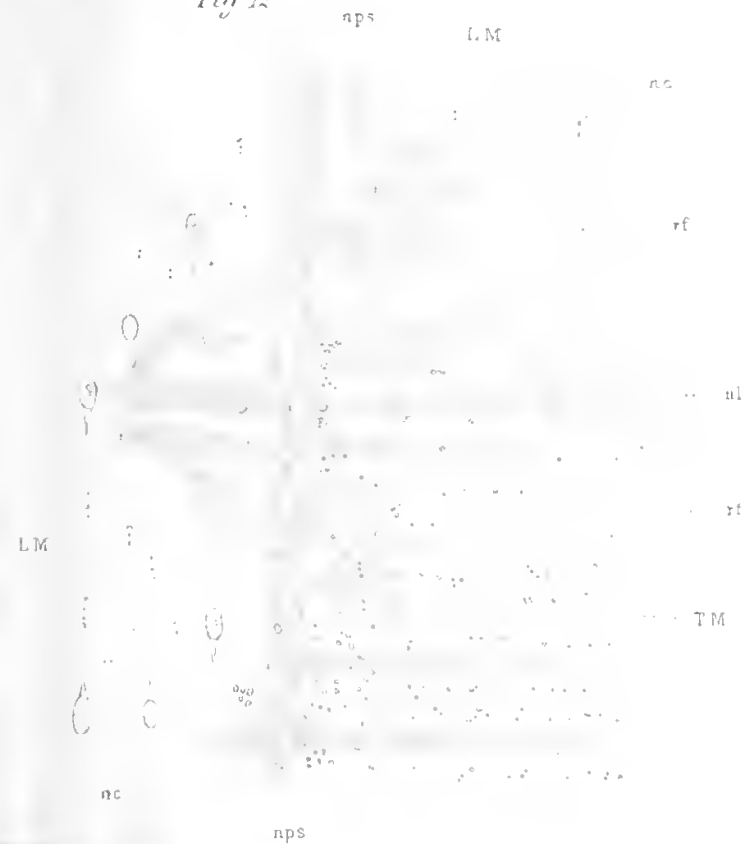
EXPLANATION OF PLATES XXXII & XXXIII,

Illustrating Dr. A. A. W. Hubrecht's Memoir on " The Peripheral Nervous System in Palæo- and Schizonemertini, one of the Layers of the Body-wall.''

Fig. 1.—Diagram of a transverse section of *Carinella*. The longitudinal nerve-trunks and the nervous sheath (indicated by red) are wholly *outside* the muscular body-wall, into which processes are being sent out by the the sheath.

Fig. 2.—The same of one of the Schizonemertini. The nervous tissue is enclosed *within* the muscles of the body-wall.

Fig. 3.—The same of one of the Hoplonemertini. The longitudinal trunks lie *inside* the muscular body-wall. The nervous sheath has disappeared, and is replaced by metameric branches placed at regular intervals and double, one being for the dorsal, the other for the ventral half. These branches divide dichotomously.

Fig. 4.—Part of a transverse section of *Carinella annulata* in the posterior half of the body. M. The longitudinal muscles of the body-wall, to which a thin layer (*m*) of circular muscles is exteriorly applied. B. Basal membrane of the skin. E. The deep cellular layers of the ectoderm, with glandular cells (*c. c*). N. The longitudinal nerve-trunk, with the inferior coating of ganglion-cells (*nc.*) applied to it, which gradually pass into the nervous sheath (*nl.*), the latter sending out processes (*np.*) into the muscular body-wall. In this figure the numerous radial fibres traversing the basal membrane (B) have been purposely omitted. Drawn with immersion vii, Seibert Krafft.

Fig. 5.—Part of a longitudinal section through the region of the nervous sheath in *Carinella polymorpha*. Letters as in fig. 4. The processes (*np.*) lead toward the cells of the ectoderm, not indicated in the figure. The nervous sheath appears more split up by fibrous and muscular tissue than in the foregoing transverse section.

Fig. 6.—Transverse section through the lateral longitudinal marrow-trunk in *Cerebratulus roseus*. L. M. Longitudinal muscular layer. C. M. Circular muscular layer, separated from the former by a layer of a homogeneous structure (like the basement membrane of the skin), through which, as through the muscular layers, pass numerous radial fibres. N. The lateral trunk situated in this homogeneous layer. Internally the fibrillar structure prevails, and this fibrillar nucleus is separated by a layer of connective tissue from the ganglion-cells (*nc*). Superiorly is represented a very large ganglion-cell, which was present in this section, though they

very rarely attain this size in the lateral trunks. *nl.* Nervous layer ensheathing the body (see Plate XXIII, fig. 12, of this volume). *np.* Nervous process penetrating among the muscles.

FIG. 7.—Oblique section (transverse horizontal) through another specimen of the same species. Letters as in fig. 6. *sct.* The layer of connective tissue between the fibrillar and the cellular nerve-tissue. These two figures and the following drawn with Siebert Krafft's immersion No. vii.

FIG. 8.—Part of the nervous sheath in a transverse section of a large specimen of *Cerebratulus marginatus.* Letters as in figs. 6 and 7. The radial fibres are omitted in this figure.

FIG. 9.—Longitudinal section through the brain and the nervous sheath of *Cerebratulus urticans,* to show the way in which the latter abuts upon the former. B. Brain-lobes. *nl.* Nervous layer. P. Outline of section through the proboscis. C. M. Circular muscular layer. L. M. Outer longitudinal muscular layer. I. M. Inner longitudinal muscular layer.

FIG. 10.—The same for *Langia formosa.* B. Superior and posterior brain-lobes. N. Lateral nerve, obliquely cut. *np.* Nervous process penetrating into the longitudinal muscles.

FIG. 11.—Part of the fibrillar nerve-sheath (*nl.*) with ganglion-cells of the foregoing preparation more strongly magnified. The process (*np.*) is seen to have essentially the same character as the fibrillar layer itself, and to be provided with several nerve-cells as well.

FIG. 12.—The nerve-sheath seen from above in a horizontal section through the back of *Cerebratulus pantherinus.* *nps.* Median dorsal nerve (nerve for the proboscidian sheath). *nl.* Fibrillar plexus with nerve-cells. *nc.* Peripheral processes from the nerve-sheath radially penetrating the muscular layers. *rf.* Radial fibres (cf. figs. 6 and 7).

FIG. 13.—A surface section through the longitudinal muscular sheath (L. M.) just above the nervous layer, with elliptic canals (*nc.*), more or less regularly arranged in transverse rows for the passage of the periphera processes of the nervous sheath. These processes show, also in this section a fibrillar structure, with nerve-cells imbedded in it. *rf.* Radial fibres.
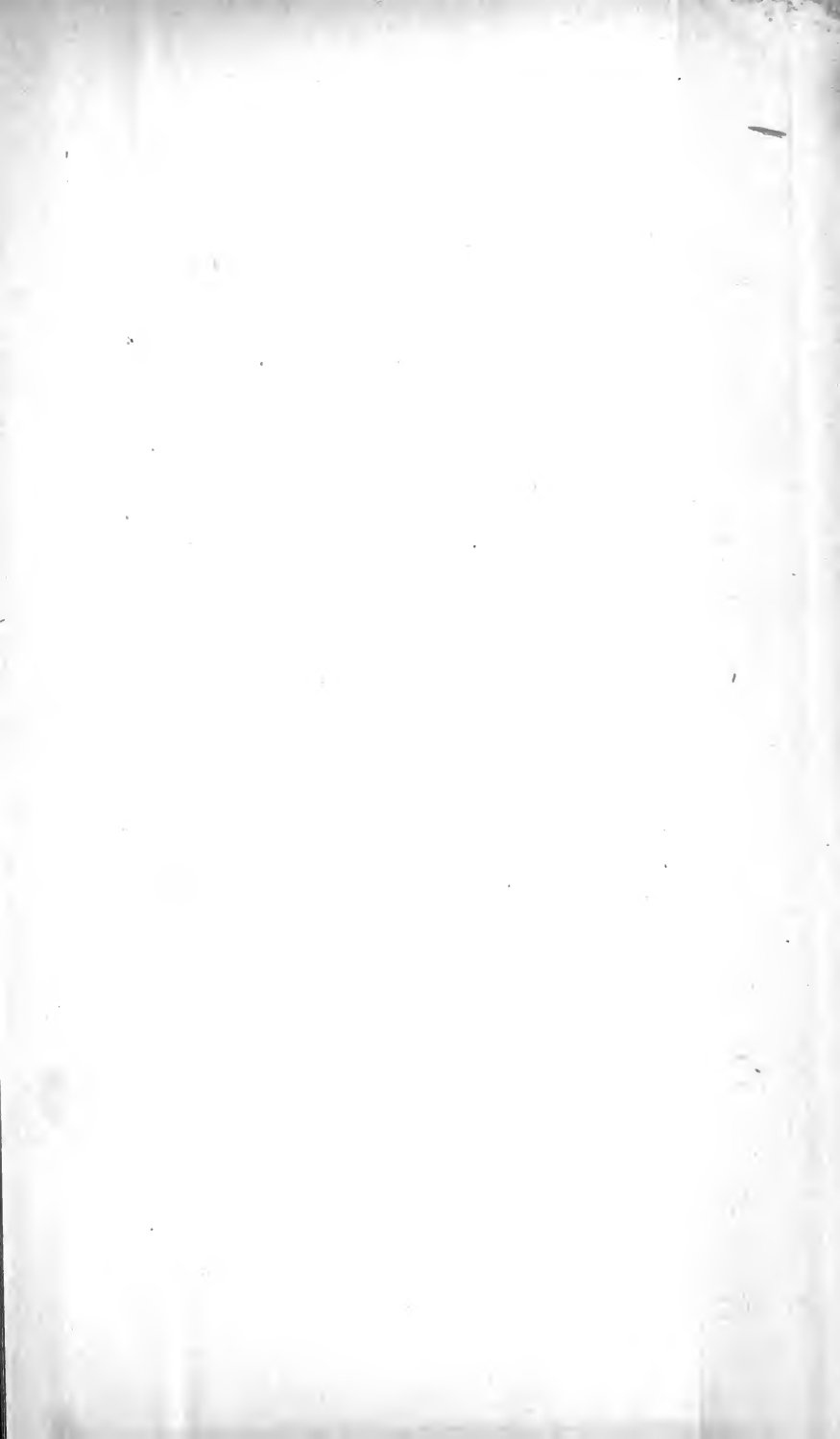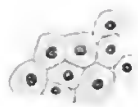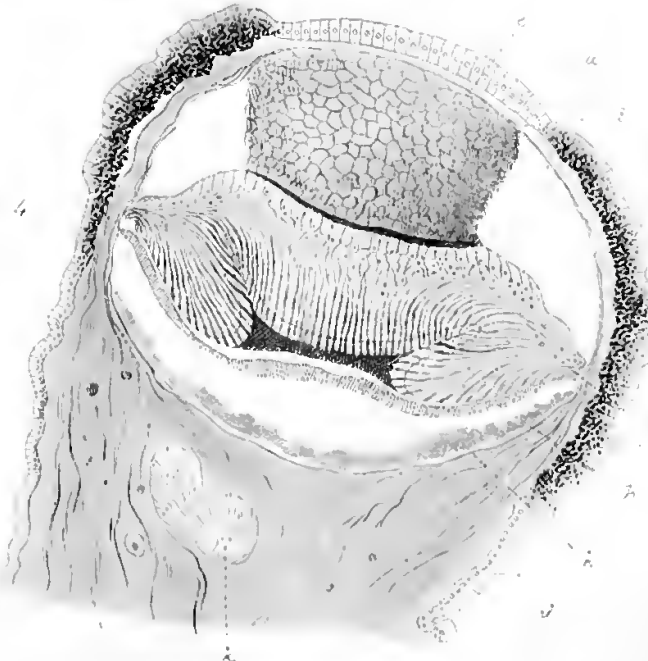
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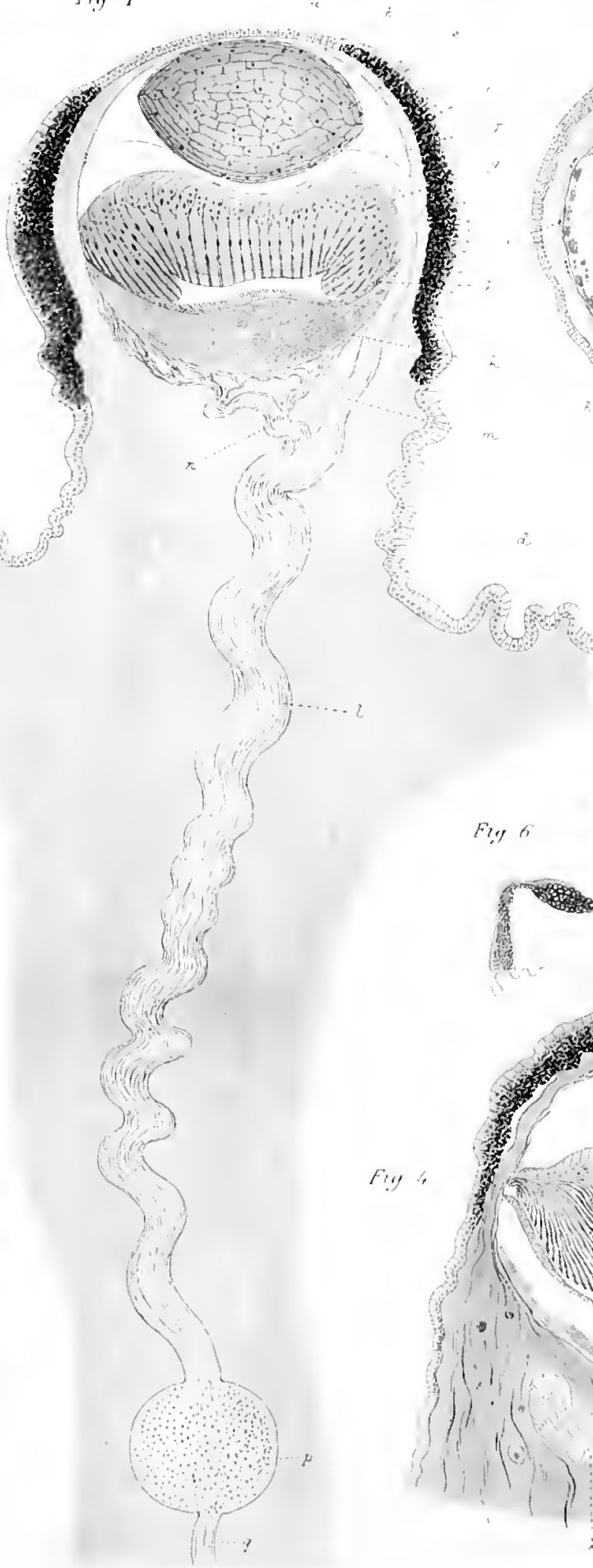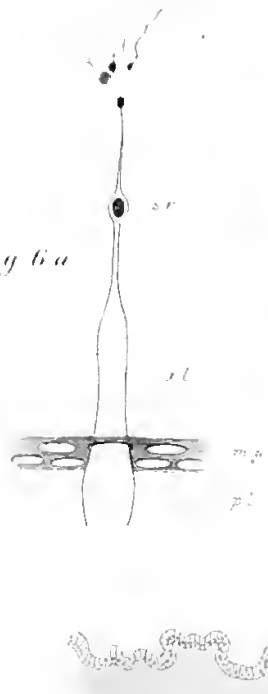
Fig 2

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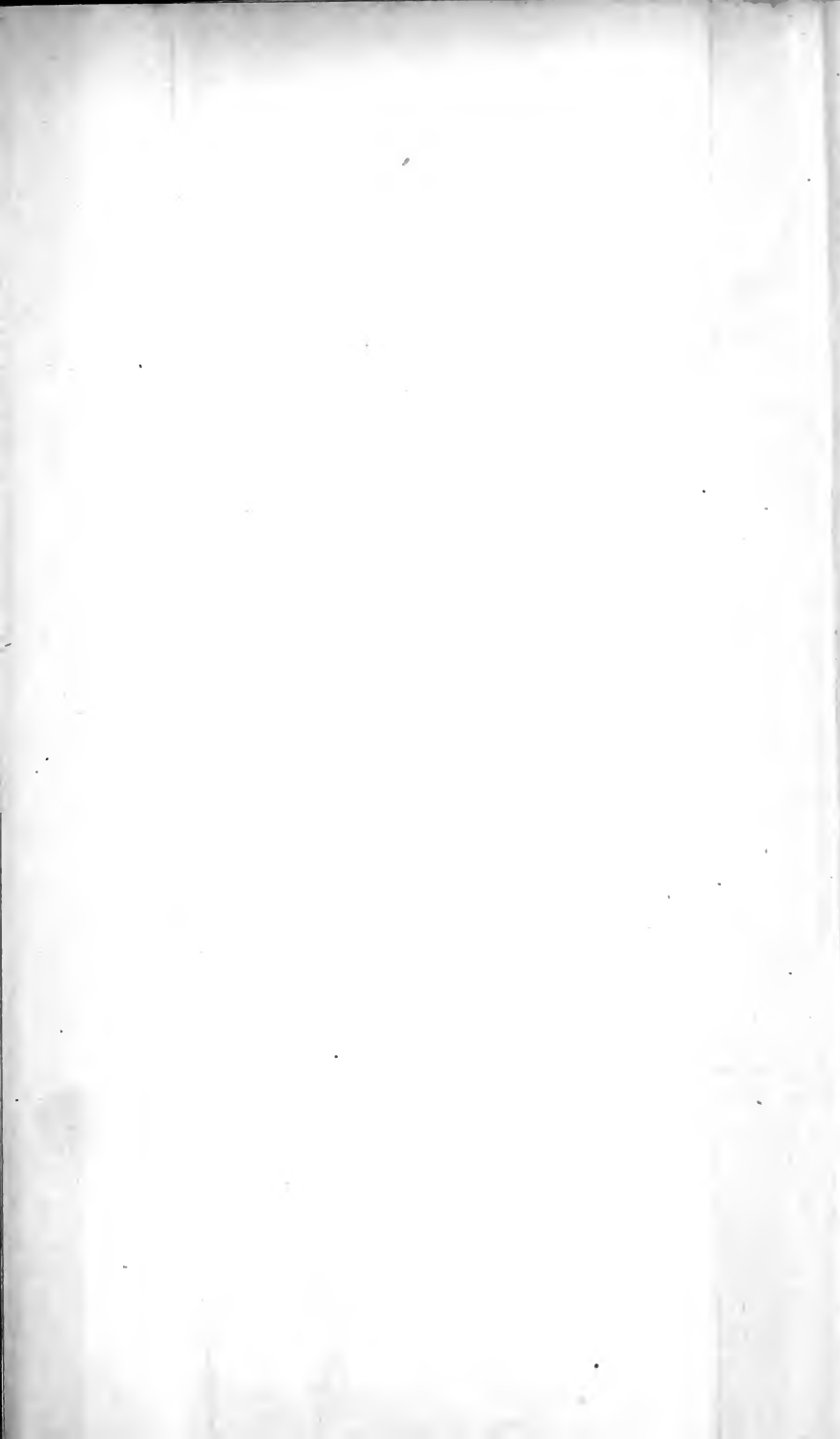
Fig. 9

Fig. 10
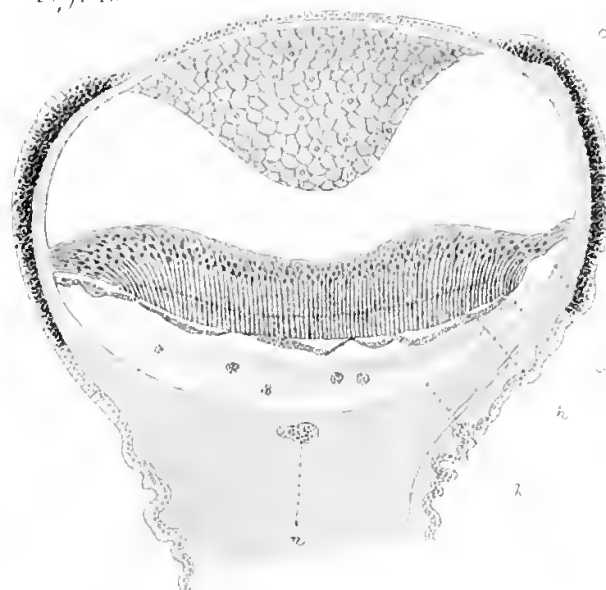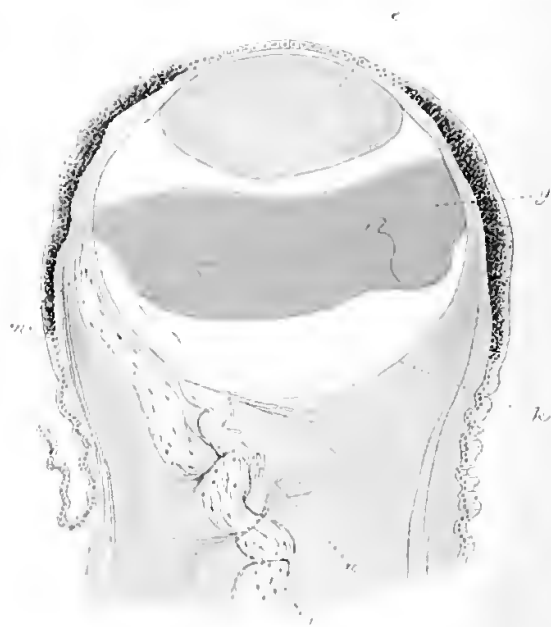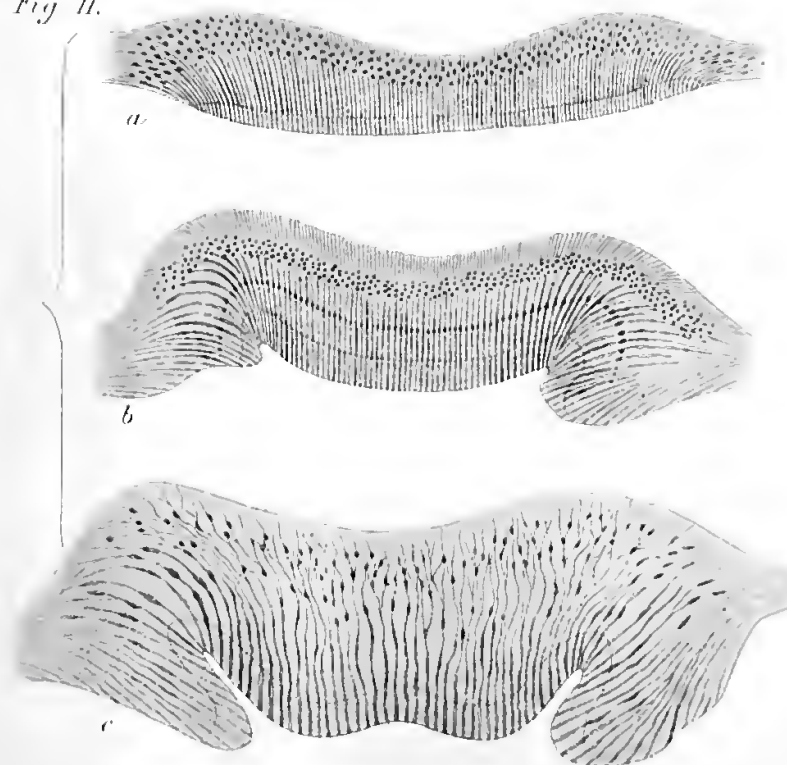
Fig. 11.

Fig. 8

EXPLANATION OF PLATES XXXIV & XXXV,

Illustrating Mr. Sydney J. Hickson's Memoir on the "Eye of Pecten."

Fig. 1.—A diagrammatic sketch of an eye of *Pecten maximus*. *a.* The cornea. *b.* The transparent basement membrane supporting the epithelial cells of the cornea. *c.* The pigmented epithelium. *d.* The lining epithelium of the mantle. *e.* The lens. *f.* The ligament supporting the lens. *g.* The retina. *h.* The tapetum. *k.* The pigment. *l.* The optic nerve. *m.* The retinal nerve. *n.* Complementary nerve. *p.* The circumpaleal nerve (Duvernoy). *q.* A supplemental nerve from pedal ganglion.

Fig. 2.—Epithelial cells of cornea.

Fig. 3.—Section through the junction of the pigmented epithelium with corneal epithelium.

Fig. 4.—Vertical section through eye of *Pecten maximus*. *a, b.* Cornea. *c.* Pigmented epithelium. *d.* Mantle epithelium. *e.* Lens. *g.* Retina. *h.* Tapetum. *k.* Pigment. *l.* Section of optic nerve.

Figs. 5, 6.—Isolated rods. 6*a.*—Diagrammatic sketch of a central rod. *p. l.* Posterior limb. *m. p.* Membrane pierced by rods. *a. l.* Anterior limb. *s. r.* Spindle-shaped portion of rod. *n.* Nerves.

Figs. 7.—Transverse sections through eye of *P. maximus*. *a.* Rods in section. *b.* Tapetum. *c.* Pigment. *d.* Retinal nerve.

Fig. 8.—Vertical section of the eye of *Pecten maximus*, showing the nerve dividing into retinal and complementary branches.

Fig. 9.—Vertical section of the eye of *Pecten maximus*, showing the termination of the retinal nerve. The retina has dropped out, and the frayed-out end of the nerve remains.

Fig. 10.—Section of eye of *Pecten opercularis*.

Fig. 11. Retina (*a*) of *P. opercularis*, (*b*) of *P. jacobæus*, (*c*) *P. maximus*.

Note.—A horizontal section means a section made in the same plane as the mantle. A transverse section, in a plane at right angles to the eye stalk. A vertical section, in a plane at right angles, both to the plane of the mantle and the last-named plane.

EXPLANATION OF PLATE XXXVI,

Illustrating Professor L. Ranvier's Memoir "On the Terminations of Nerves in the Epidermis."

Fig. 1.—Section of the epidermis of the snout of the pig, with the nervous ramifications stained by chloride of gold.

Fig. 2.—Section of an organ of Eimer on the nose of a mole, with the nerves which supply it.

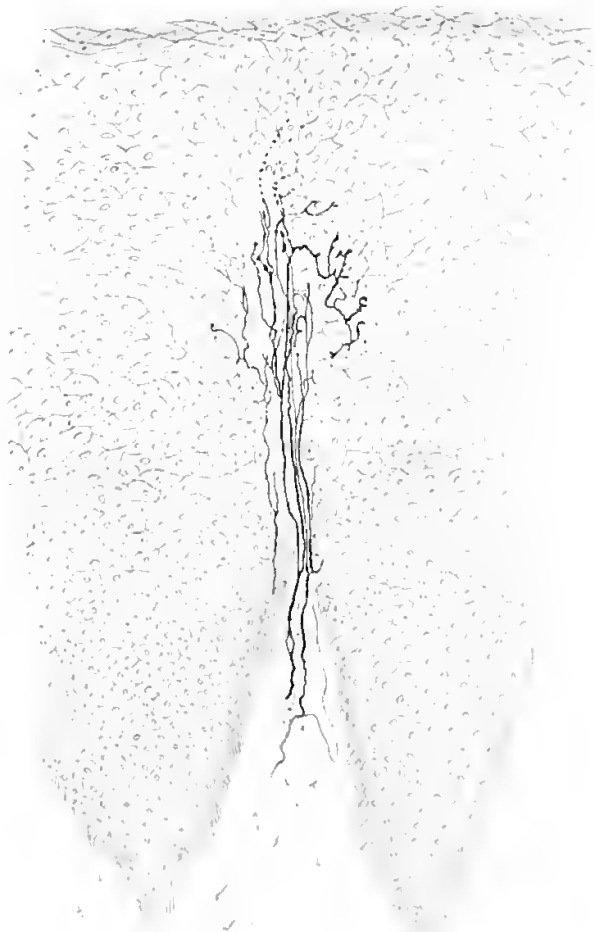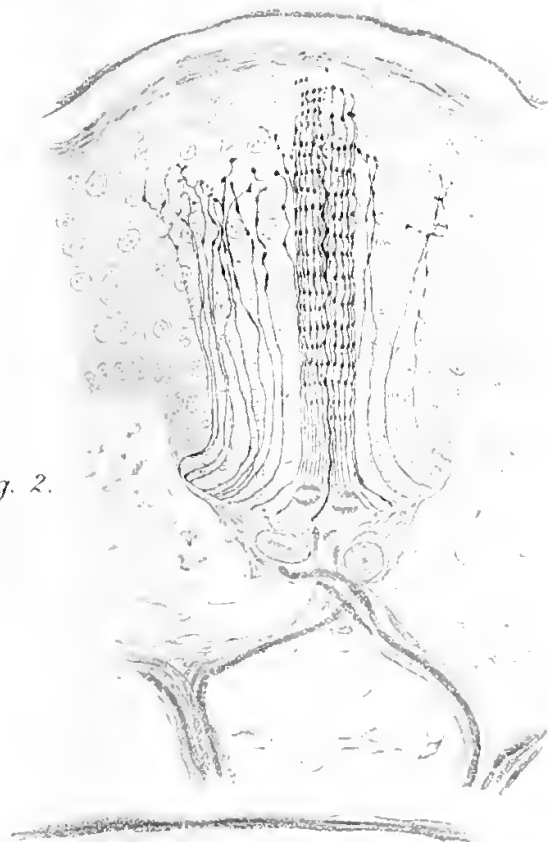Fig. 3.—Section of the epidermis of the human finger.
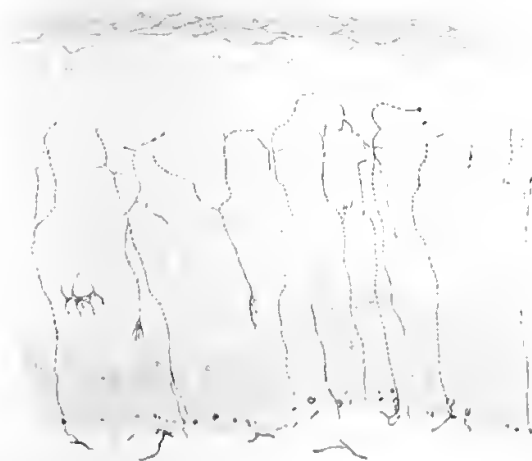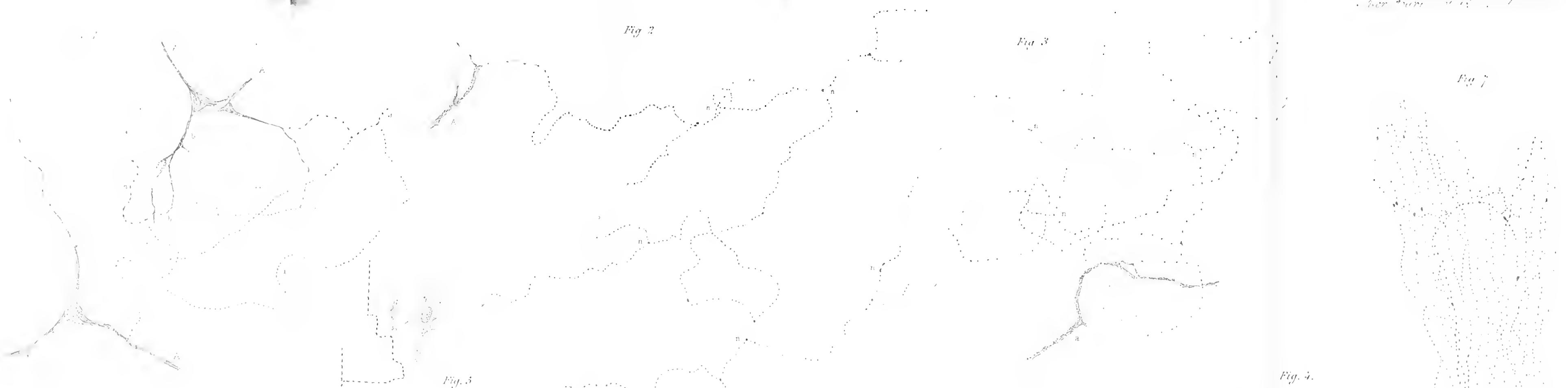
Fig. 2.

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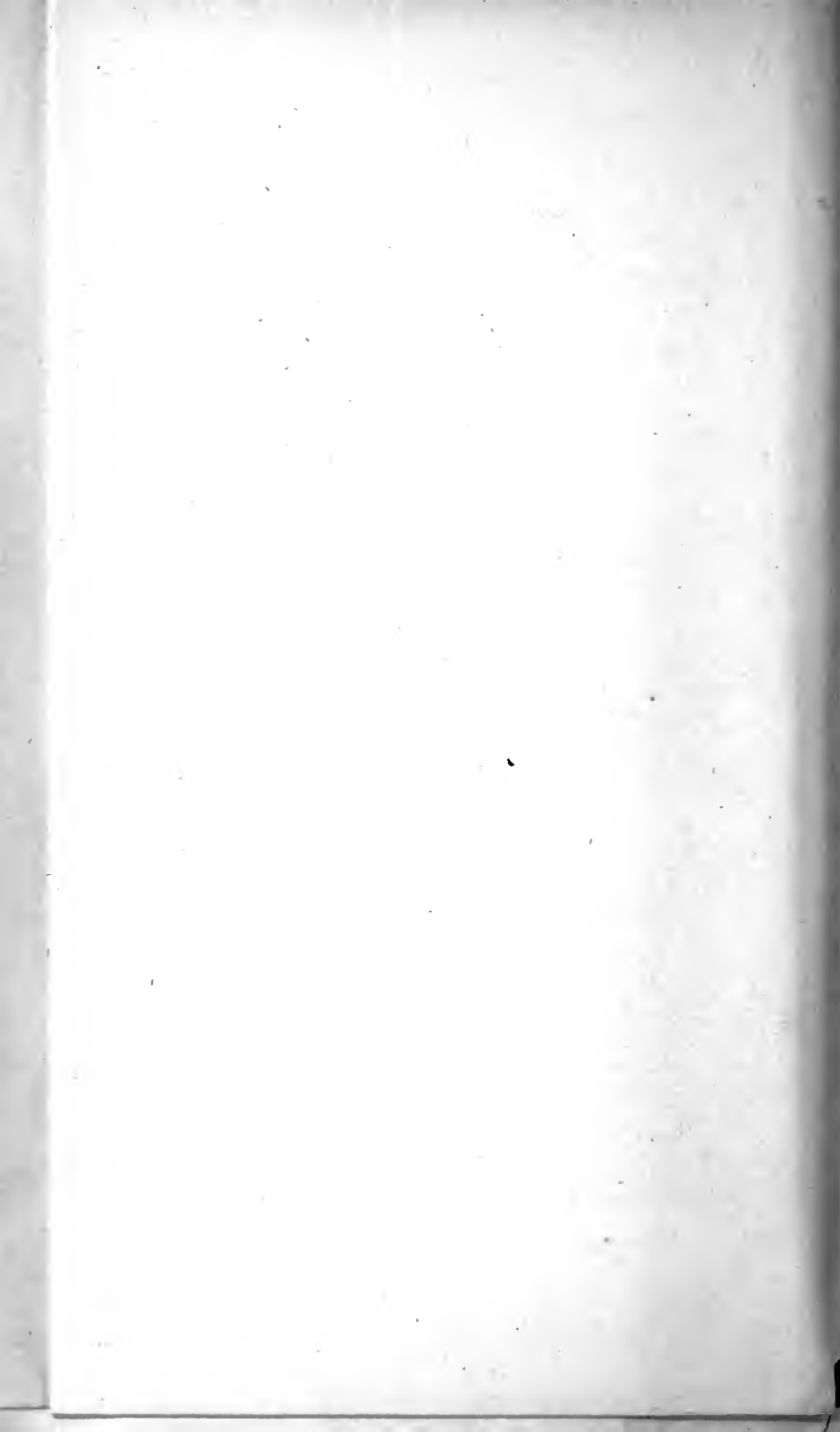
EXPLANATION OF PLATE XXXVII,

Illustrating Dr. E. Klein's Memoir on the "Termination of the Nerves of the Mammalian Cornea."

(Figures 4 and 6a are drawn by Mr. Noble Smith, all others by the author.)

Fig. 1.—From a horizontal section through the rabbit's cornea. Magnifying power about 260. Showing the subbasilar plexus proper. A. Branches of the stroma plexus, situated behind and close to Bowman's membrana elastica anterior. *a.* Fine nerve-fibrils, at first in the same level as A, but, passing into a deeper layer, terminate in the substantia propria. *b.* Thicker nerve-fibrils, somewhat anterior to A, being situated within Bowman's membrane. *x.* Fine nerve-fibrils, which enter the subepithelial network. *xx.* Bundles of primitive nerve-fibrils which come off from the rami perforantes and enter the subepithelial network.

Fig. 2.—From the same cornea as fig. 1. Magnifying power about 260. A. Branch of the stroma plexus of the same level as A in fig. A. *n.* Fine nerve-fibrils situated within Bowman's membrane, the intrabasilar fibrils. At *x* they pass into the depth again, and finally terminate in the substantia propria. At *xx* they enter the subepithelial network.

Fig. 3.—From the same preparation as fig. 2. Magnifying power about 260. *a.* Branch of the stroma plexus, similar to A in the preceding figures. *n.* Fine nerve-fibrils immediately behind the subbasilar plexus of fig. 1. These are the deep subbasilar fibrils referred to in the text. Most of them terminate in the substantia propria.

Fig. 4.—From the same cornea as the preceding figures. Magnifying power about 440. Showing fine nerve-fibrils of the substantia propria of the cornea, and their relation to the corneal corpuscles. These latter are only indicated as granular plates, their processes have not been drawn. At *a* apparent terminations.

Fig. 4a.—From a similar cornea as in the preceding figure. Magnifying power about 660. Showing the relation of the fine nerve-fibrils of the substantia propria to the corneal corpuscles. At *a* apparent terminations.

Fig. 5.—From a cornea of a kitten. Magnifying power about 660. Showing the relation of a fine nerve-fibril to the processes of a corneal corpuscle. This latter being stained of a grey colour, and the nerve-fibril black, the distinction between the two is easily made. The nerve-fibril is not actually connected with the processes of the corneal corpuscle, but lies close to them, both being contained in the lymph-canalicular system. This applies equally to figs. 4 and 4a.

Fig. 6.—From the same cornea as in the preceding figure. Magnifying power about 1400. Showing a very fine nerve-fibril giving off a lateral branchlet, which terminates *on* not *in* the corneal corpuscle in a fine network.

Fig. 6a.—From the cornea of a frog. Magnifying power about 660. At *a* are shown the ultimate fine nerve-fibrils forming a network on the corneal corpuscle. At *b* an apparent termination, the ultimate fibrils are not brought out in the preparation.

Fig. 7.—Subepithelial network of fine nerve-fibrils of the cornea of

rabbit seen in a horizontal section. Magnifying power about 660. 1 and 2 are two bundles of fine nerve-fibrils coming off separately from two rami perforantes, not included in the drawing. As is shown in the drawing, not only are there anastomoses between fibrils of the same bundle, but also, as at 3, between the fibrils of the neighbouring bundles.

Fig. 8. From a similar preparation, showing the fibrils of the subepithelial network in a horizontal section. Magnifying power about 440. All these fibrils, like those of the preceding figure, run horizontally close behind the deep layer of the anterior epithelium of the cornea. The actual length of the part of the preparation depicted here is 0·4 mm., and all the relations of the individual fibrils and their branchlets are drawn with great accuracy, in order to show the relative lengths of the fibrils of the subepithelial network, and the number of lateral branchlets which pass into the anterior epithelium. A, B, C are three bundles of fine fibrils coming off from separate rami perforantes. In the bundle A a thick fibril can be followed for a very great length. Of the fibrils with which it anastomoses *f* and *g* are noteworthy on account of their length, and on account of the numerous short fibrils, given off by them, which enter the anterior epithelium at *x* ; *x* indicates all those fibrils which ascend into the anterior epithelium. *k*. A nerve-fibril, through which the fibrils of neighbouring bundles anastomose with one another.

Fig. 9.—From a horizontal section through the cornea of guinea-pig. Magnifying power about 440. Showing the nature and mode of division and anastomoses of the intraepithelial nerve-fibrils in the superficial layers of the anterior epithelium. The very minute rod-like lateral branchlets are very conspicuous. At *n* a closed network.

Fig. 10.—From a horizontal section through the cornea of rabbit. Magnifying power about 660. Showing the intraepithelial nerve-fibrils, and the character of their distribution. At F a very fine fibril. The numerous minute lateral branchlets are well shown. At *a*, *b*, *c* there is a closed network. In the upper right part of the drawing is an intraepithelial branched corpuscle shown. It has no connection with the nerve-fibrils.

Fig. 11.—From a horizontal section through the cornea of rabbit. Drawn with the $\frac{1}{12}$ oil immersion of Zeiss. Showing the intraepithelial nerve-fibrils, their varicosities, branching, anastomoses, and terminations.

a and *b*. Two nuclei of superficial epithelial cells, to show the relative proportions. *c*. The terminal network seen in profile between two epithelial cells. *d*. The terminal network seen *en face*.
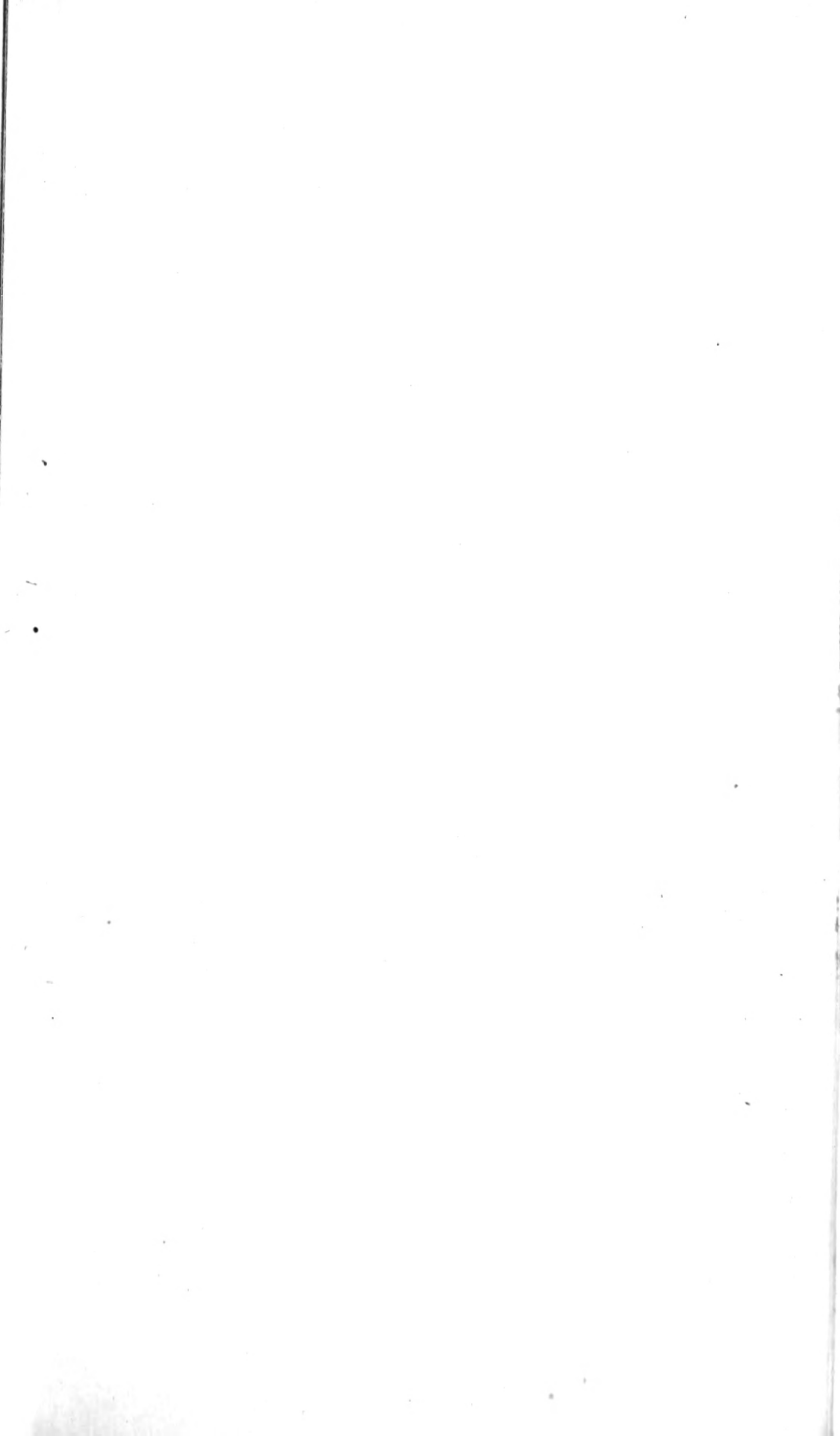
Fig. 12.—Intraepithelial nerve-fibrils in a horizontal section through the cornea of rabbit. Magnifying power about 440. The numerous minute rod-like branchlets are shown here.
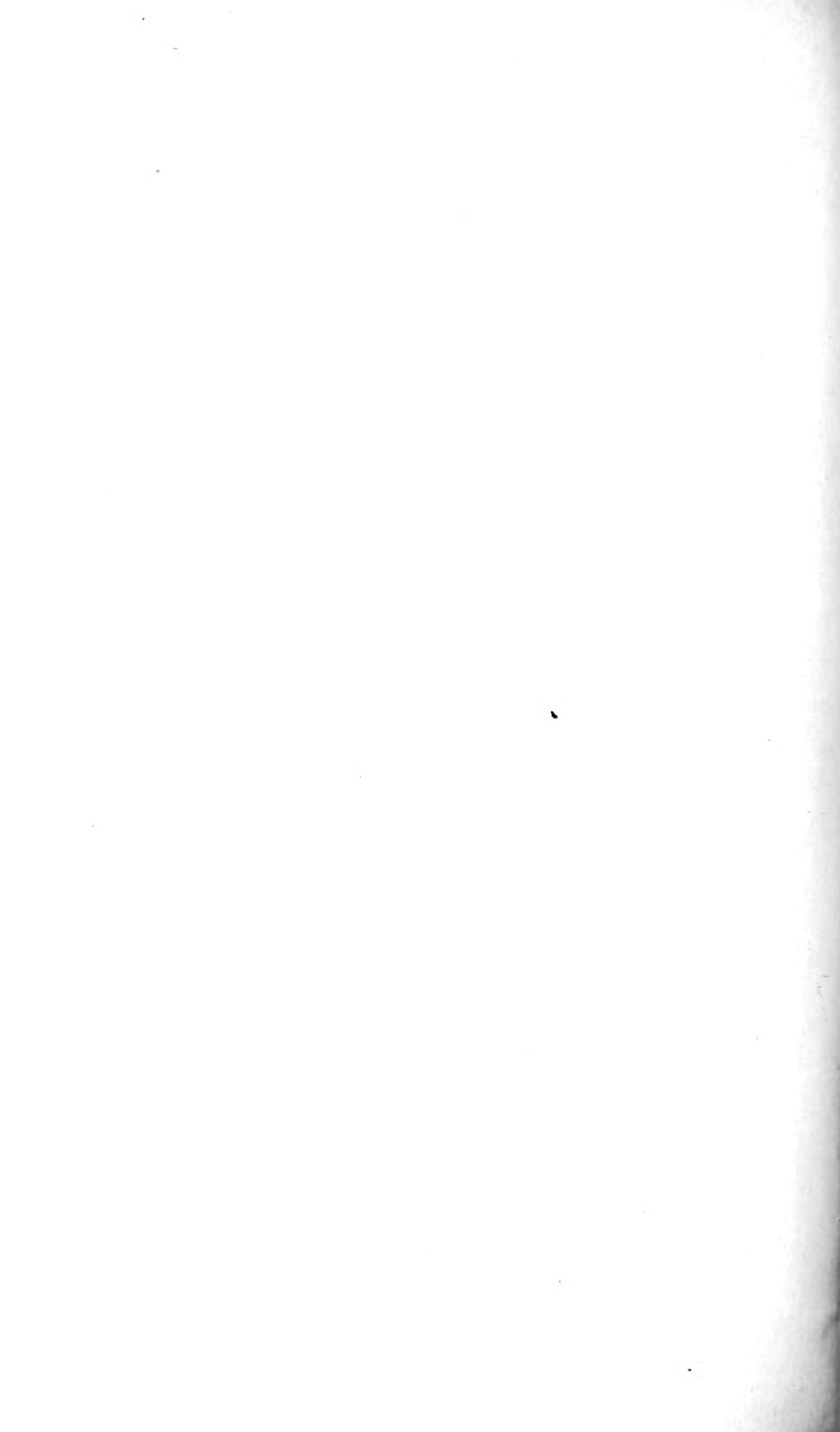
Fig. 13.—From the same cornea. Magnifying power about 1020. Showing some of the very fine superficial intraepithelial nerve-fibrils, their minute branchlets just indicated.
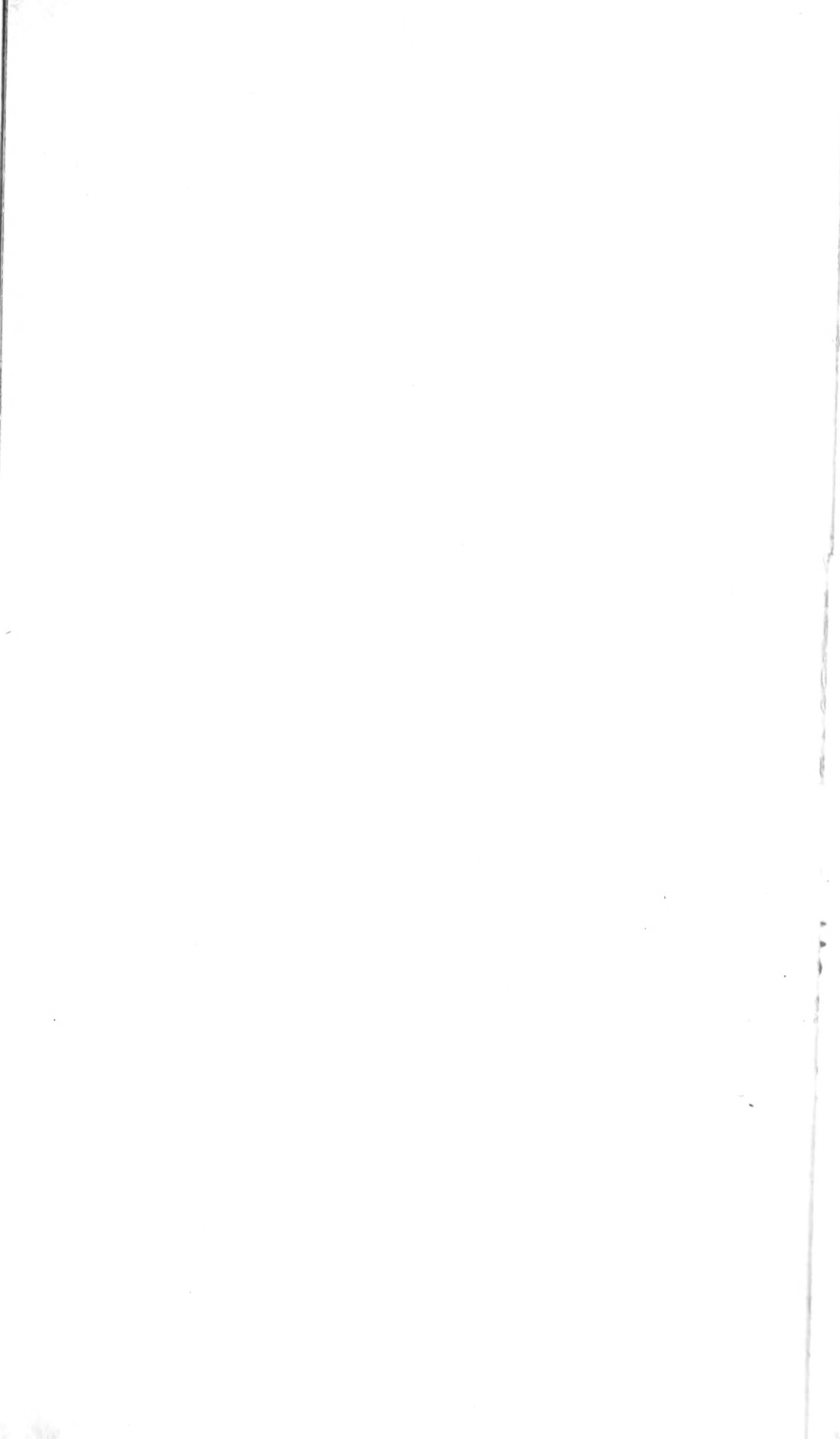
Fig. 14.—From the same specimen. Magnifying power about 660.

Fig. 15.—From the same specimen. Magnifying power about 1020. In both figures the fine intraepithelial nerve-fibrils of the superficial layers are shown, and their ultimate networks as seen from the surface. The oval corpuscle in either figure represents the nucleus of an epithelial cell.

Fig. 16.—From a similar specimen. Drawn with Zeiss's $\frac{1}{15}$ oil immersion. Showing the terminal network of the fine nerve-fibrils. The oval corpuscles represent two nuclei of superficial epithelial cells, to show the relative proportions. The terminal network is in no connection with the nuclei.